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NOTES ON CERTAIN ASPECTS IN THE BIOLOGY OF THERAPON PLUMBEUS (KNER)

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TEN TEXT FIGURES

FOREWORD

The silvery theraponid, *Therapon plumbeus* (Kner) is considered one of the commercially important fresh-water fishes, it being caught in considerable quantity from Laguna de Bay, where it abounds in all portions of the lake. It is fairly small, seldom exceeding a length of 150 mm; quite bony but nontheless possessing fairly good flavor. As such it is fairly priced and is thus very popular and in big demand among the poorer classes of people. It is available practically throughout the year; it is a good substitute for the other more favored but seasonally caught fishes.

One of man's sources of food, it also serves as forage for larger fishes in the lake. Too, it is used as bait in catching carnivorous fishes.

Since the species is available in abundance almost the year round, it can be fairly assumed that its exploitation do contribute materially to the income of small scale fishermen of Laguna de Bay. Being a good source of food fish for a great many people around the lake, it carries the implication that the species should be so managed to insure a maximum sustained yield (in lbs or Kg) in the years to come. In this

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respect, there are several factors considered germane to proper management of the fresh-water population. Among these are: knowledge of (*1*) their enemies; (*2*) their rate of growth; (*3*) fecundity or productiveness; (*4*) spawning season and life history, a must in all conservation works. Having these objectives in mind and considering that the species looms large in the fisheries of Laguna de Bay, a certain knowledge of its biology becomes not only proper but necessary. This paper, therefore, aims to supply that certain information on the biology of the fish, specifically with respect to spawning season, fecundity and possible age determination through the length-frequency distributions.

DISTRIBUTION

The silvery theraponid or *ayuñgin* as the fish is locally known is widely distributed in the island of Luzon; it is a fresh-water form, colonizing rivers, lakes, fishponds and even rice paddies. In Laguna de Bay it abounds in practically every portion of the lake. Herre's report (1930) on this fish and later concurred in by Villaluz (1953) listed the fish as one of the indigenous fresh-water fishes of the Philippines.

NOMENCLATURE

Umali (1936) listed the theraponids under the family Theraponidæ, order Percomorphi. His work listed nine species of *Therapon*. Only the silvery theraponid is fresh-water form; all others are marine. In his check list of Philippine fishes, Herre (1953) considers it as *Datnia plumbea*, a name given by Kner in 1865 after he collected and described the fish. However, Bleeker (1873) used the genus *Datnia* as synonym to *Therapon* and gave the name *Therapon* (*Datnia*) *plumbeus*. Fowler (1927) used the following genera for the same species: *Mesopristes* (1918), *Therapon* (1927), and *Terapon* (1931).

Neave (1939) in his "Nomenclator Zoologicus" showed clearly that while the genus *Datnia* was first used by Cuvier and Valenciennes only in 1829, the genus *Therapon* was already used by Cuvier in 1817 and Cloquet in 1819. Thus is established the priority of the genus *Therapon* over *Datnia*, hence the fish should be *Therapon* (*Datnia*) *plumbeus* (Kner).

REVIEW OF LITERATURE

As far as is known locally not much work has been done on the study of the silvery theraponid. Seale (1908) included

this species in his list of the chief food fishes in Philippine waters. Aldaba (1931) in his work on the different fishing methods in Laguna de Bay stated that it is one of the few species caught in considerable quantity. Mane (1934) worked out its spawning and feeding habits and found that the fish spawn almost the year round as evidenced by the findings that females had ovaries showing eggs at various stages of maturity. The present paper confirms this findings. Mane (op. cit.) showed, likewise, that the peak of spawning activity occurs during the dry season; namely, in the months of March, April, and May. Activity slows down gradually during the months of June to December only to resurge in January to May. It was also shown that *Therapon* is omnivorous, subsisting on both plant and animal food. Roxas and Martin (1937) contributed a systematic account of the fish. Villaluz (op. cit.) listed the silvery theraponid as one of the true indigenous fresh-water fishes of the Philippines.

METHODS

Collection of materials.—The material used in this study consists of collections of silvery theraponids caught by means of cast nets for a period of one year, commencing December, 1957 and ending November, 1957. All catches were done in Tanay, a town of Rizal Province located on the eastern shore of Laguna de Bay. Samples, for the most part, were made weekly except for the month of February when only one sample was taken; August, two samples. For January, June and September, three sample each month were taken. The collection for any one month was never less than one hundred specimens, and the total number of fish collected for the period under study amounted to 2,528 (Table 1).

Age determination.—An attempt was made to determine the age of the fish based upon length-frequency distributions. The total length (from the tip of the snout to the tip of the longest caudal fin ray) of each fish was measured and recorded to the nearest millimeter. The fish were then divided into groups of 5 mm intervals (Table 2). Since the number of fish measured is not constant for each month, then the length frequency distributions were made comparable by converting the frequencies into percentages of the total for the month and then the data smoothed out once by a moving average of 3.

TABLE 1.—*Summary of fish collections from December, 1956 to November, 1957.*

Date of collection	Males	Females	Total	
			Per sample	Per month
December 2, 1956	5	32	37	
December 9, 1956	52	51	103	
December 16, 1956	47	14	61	
December 27, 1956	36	9	45	246
January 6, 1957	12	17	29	
January 13, 1957	0	47	47	
January 25, 1957	40	12	52	128
February 23, 1957	65	43	108	108
March 2, 1957	34	40	74	
March 9, 1957	26	31	57	
March 16, 1957	39	42	81	
March 23, 1957	48	31	79	292
April 2, 1957	28	43	71	
April 11, 1957	29	51	80	
April 23, 1957	36	50	86	
April 30, 1957	51	39	90	327
May 7, 1957	20	21	41	
May 14, 1957	60	12	72	
May 21, 1957	47	20	67	
May 28, 1957	27	9	36	216
June 4, 1957	23	5	28	
June 10, 1957	62	13	75	
June 17, 1957	27	31	58	171
July 1, 1957	43	11	54	
July 8, 1957	37	12	49	
July 19, 1957	62	27	89	
July 28, 1957	41	11	52	244
August 21, 1957	67	23	90	
August 26, 1957	30	15	45	135
September 1, 1957	89	70	159	
September 15, 1957	21	23	44	
September 23, 1957	29	34	63	216
October 7, 1957	35	21	56	
October 14, 1957	29	17	46	
October 20, 1957	36	7	43	
October 27, 1957	35	15	50	195
November 3, 1957	48	22	70	
November 10, 1957	34	15	49	
November 17, 1957	26	25	51	
November 25, 1957	10	40	50	220
Total	1,587	941		2,528

The results are plotted in Figures 1, 2, and 3. On the right hand side of each monthly length-frequency distribution are indicated the total number of specimens examined.

Also a study of the relation between length and weight was made based on the measurements in total length and the weight in grams of 504 specimens, 247 females and 257 males. The fish were grouped in 5 mm classes, the average weight of the fish for each class taken (Table 3) and then data was plotted on ordinary graphing paper. From the data obtained a length-weight curve was fitted using the method of least squares. Goodness of fit of the curve was verified by the Chi-Square test. The formula used was the exponential equation $W = aL^n$, where W represents the weight expressed in grams; L the total length

TABLE 2.—*Length frequency table of 2,528 silvery theraponids collected from December, 1956 to November, 1957.*

Class interval	December, 1956			January, 1957			February			March			April			May			June			July			August			September			October			November			Total		
	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M	F	M	F and M
50–54																	1	1																		1		1
55–59		4	4		2	2							1	1	2	2	4	6		1	1				1		1							4	12	16
60–64	3	13	16	3	7	10							1	1	2	4	9	13	2	3	5	2	3	5		2	2	3	2	5	1	2	3	1	1	2	21	42	63
65–69	17	16	33	4	20	24	1	2	3	2	10	12	6	6	12	3	7	10	2	2	4	2	6	8	3	5	8	13	12	25	3	7	10		2	2	68	97	165
70–74	11	25	36	3	22	25	6	14	20	6	11	17	8	10	18	3	8	11	1	3	4	5	13	18	11	23	34	12	15	27	9	19	28	5	8	13	89	177	266
75–79	19	33	52	1	13	14	8	11	19	7	12	19	9	15	24	4	7	11	1	5	6	5	13	18	1	9	10	11	15	26	6	13	19	9	16	25	85	130	215
80–84	18	14	32		9	9	7	12	19	8	9	17	7	16	17	7	12	19	6	13	11	5	6	11	6	7	13	21	20	41	5	11	16	5	15	20	97	174	271
85–89	14	11	25	2	10	12	3	8	11	12	20	33	8	17	25	7	25	32	11	27	38	5	13	18	2	13	15	20	12	32	11	20	31	16	17	33	132	206	338
90–94	5	11	16	6	10	16	3	7	13	20	25	45	24	29	53	4	28	32	3	31	34	12	26	38	4	5	9	11	8	19	9	19	28	17	17	34	119	230	349
95–99	7	7	14		3	3	3	6	9	18	18	36	28	55	83	6	28	34	3	28	31	2	26	28	2	12	14	11	5	16	6	15	21	9	22	31	94	182	276
100–104	1	3	4	2	1	3	1	3	4	25	22	47	20	30	50	2	15	17	4	8	12	2	19	21	1	2	3	5	6	11	4	7	11	6	6	12	57	86	143
105–109		1	1	2	3	5	1		1	16	11	27	10	14	24	6	9	15	3	6	9	1	9	10		1	1	1	3	4	2	3	5	3	5	8	37	43	70
110–114	1		1		1	1	1		1	7	3	10	7	5	12	1	7	8							1	1	2	1	1	2		2	2	12	2	14	29	25	54
115–119				1	2	3	1		1	7	6	13	3	2	5	1	1	2	1	1	2	1	1	2	1	1	2	1		1		1	1	1	7	8	17	9	26
120–124	1		1		1	1	1		1	5	3	8				1		1		1	1	1	2	3				1		1				1	2	3	6	7	13	4	17
125–129	1		1							3		3																									7	5	12
130–134	1	1	2					1	1	3	1	4																									2		2
135–139	1		1							1		1																								1		1		
140–144	2		2																										1	1	2							1	1	2
145–149																																					1		1
150–154	1		1																																				
155–159																																							
Total	106	140	246	24	164	188	43	65	108	141	151	292	132	195	327	62	154	216	45	126	171	61	183	244	88	97	185	127	119	246	60	135	195	102	118	220	941	1,587	2,528

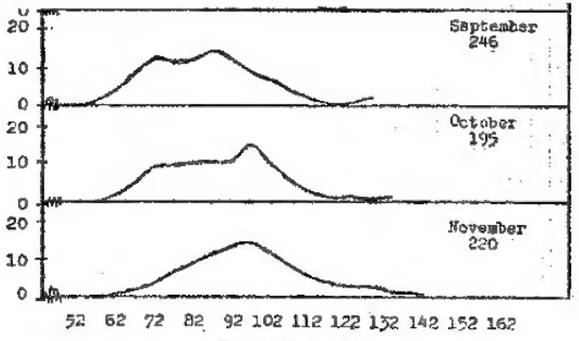

FIG. 1. Monthly length-frequency polygons of silvery theraponids
Sexes combined.

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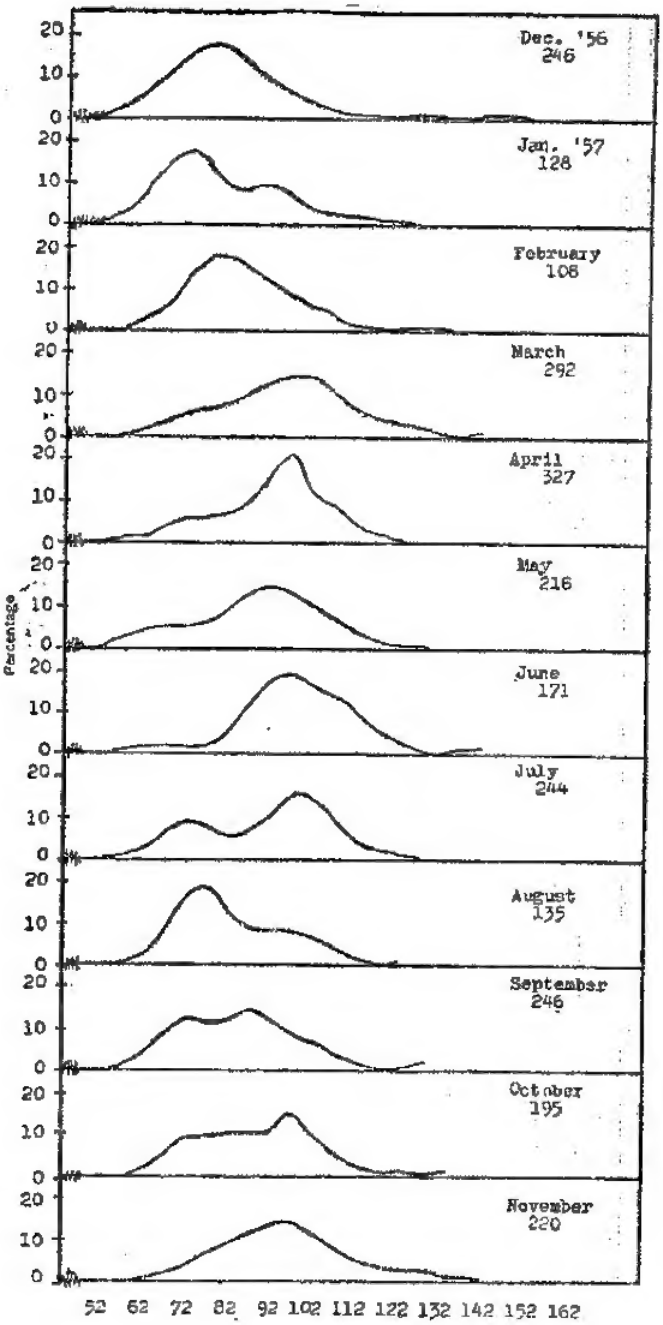

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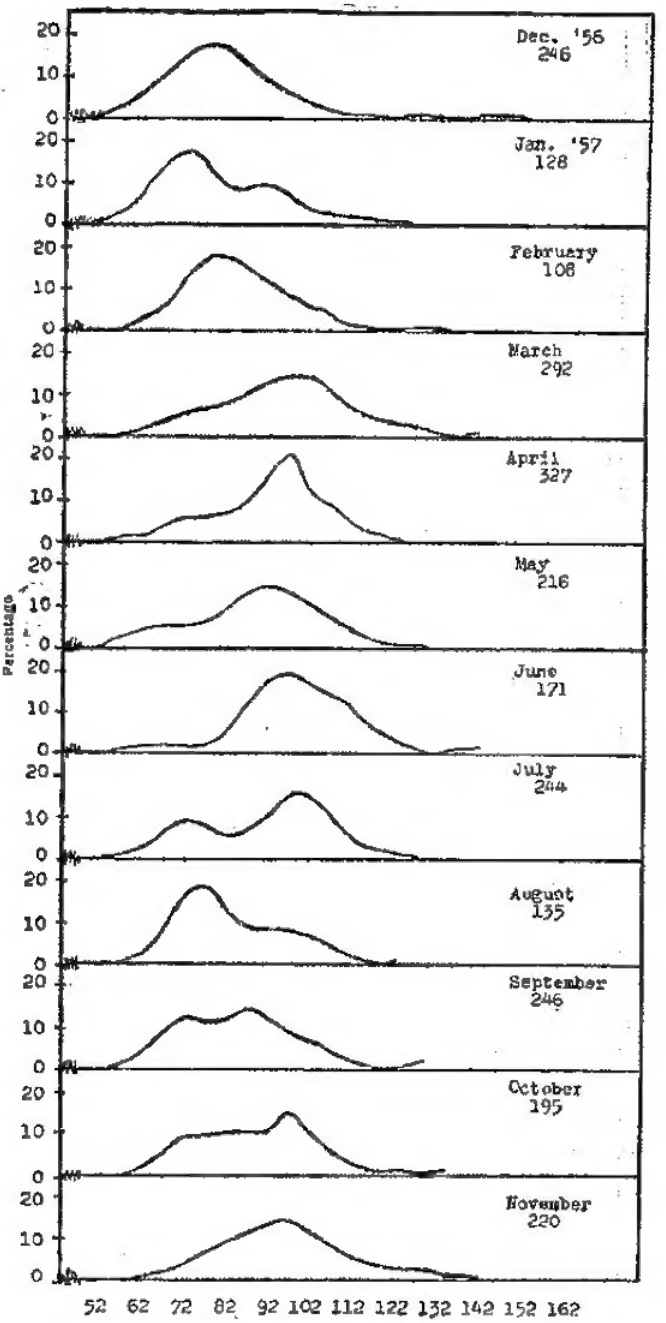

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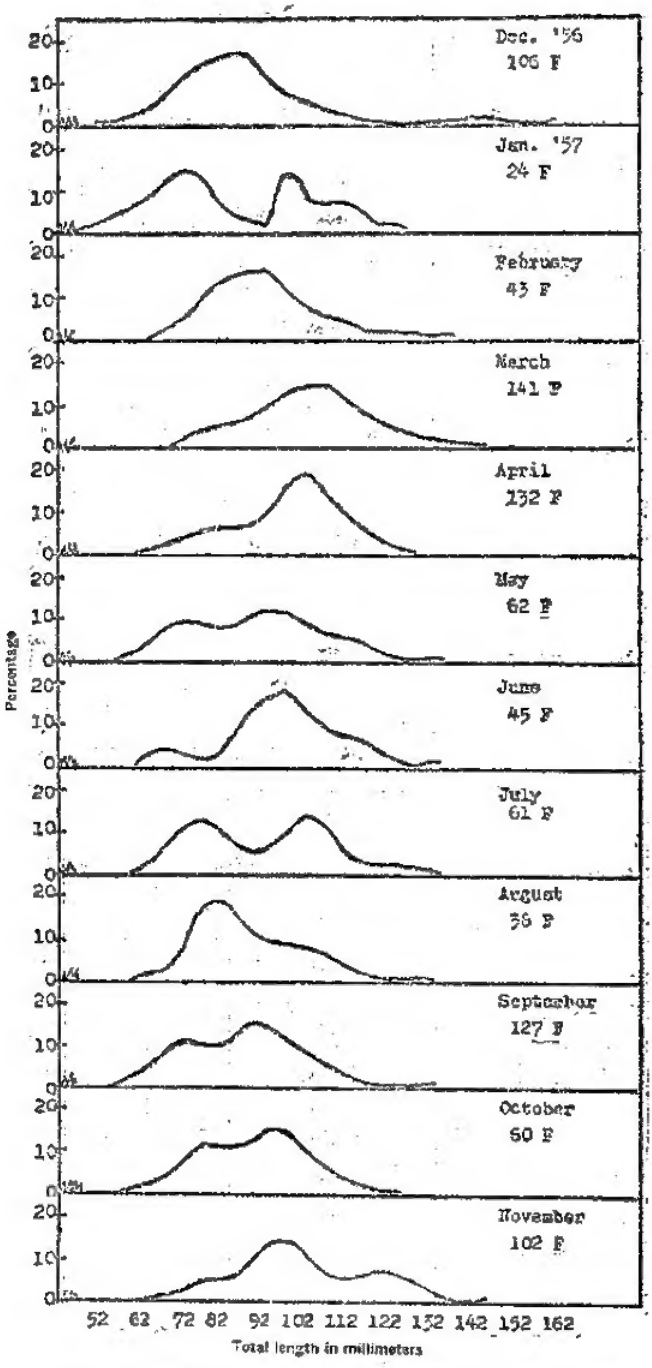


FIG. 2. Monthly length-frequency polygons of female silvery theraponids.

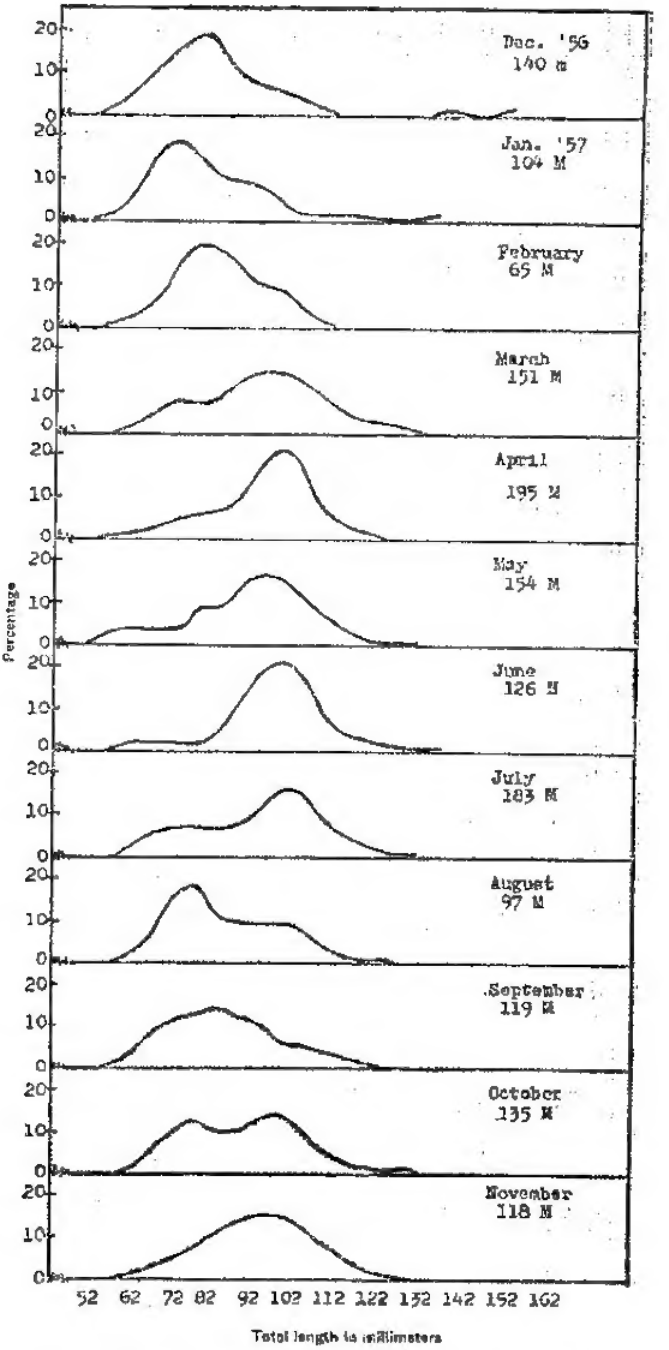


FIG. 3. Monthly length-frequency polygons of male silvery theraponids.

TABLE 3.—*Average weights in grams of 247 females and 257 males for every group length (in mm).*

Total length in (mm)	Average weights in grams		
	Females	Males	Both sexes
57	2.88	2.37	2.50
62	3.27	3.10	3.18
67	3.29	3.58	3.47
72	3.92	4.75	4.20
77	5.09	5.16	5.13
82	6.45	6.74	6.60
87	7.90	8.51	7.01
92	10.21	9.94	10.07
97	11.70	12.78	12.60
102	14.40	14.30	14.36
107	17.47	17.14	17.23
112	20.75	20.32	20.59
117	23.13	22.89	23.03
122	28.03	24.11	26.44
127	30.55	26.66	29.58
132	32.68	32.80	32.71
137	36.85	----------	36.85
142	40.90	----------	40.90
147	44.40	----------	43.85

in millimeters; a and n are the constants. The formulæ in the determination of the values of a[1] and n[2] are:

$$\log a = \frac{\Sigma \log W. \Sigma (\log L)^2 - \Sigma \log L. \Sigma (\log L. \log W)}{n. \Sigma (\log L)^2 - (\Sigma \log L)^2}$$

$$n = \frac{\Sigma \log W - (n. \log a)}{\Sigma \log L}$$

Spawning season.—It is quite difficult to distinguish between sexes in the fish due to the absence of sexual dimorphic characters. Specimens were, therefore, opened up to determine the sex and also to determine the degree of maturity of the fish. The latter determination is accomplished by the examination of ovaries and the measurement of the diameter of ovarian ova in the various stages of development. Male specimens were considered only for the determination of sex ratio.

The ovaries from each female were carefully removed from the fish, the length measured and weight taken for fecundity studies. The ovaries were opened up and examined with the used of a low power binocular microscope to determine the degree of maturity using the following stages as criteria:

Stage I. Immature; the ovaries thin and elongated with compact transparent walls; eggs not visible to the naked eye; eggs totally transparent under the binocular, polygonal in shape due

[1] Lagler, K. F., Fresh-water Fishery Biology (1956) 165.
[2] Ibid.

to the crowding of the cells whose sizes range from 0 to 0.19 mm.

Stage II. Maturing; gonads are slender and elongated too; eggs still invisible to unaided eye; microscopic examinations show isolated, rounded cells; 0.20 to 0.25 mm in diameter.

Stage III. Maturing; gonads enlarged and granular in appearance; ova opaque, yolked, with translucent peripheral ring; 0.26 to 0.55 mm in diameter.

Stage IV. Maturing; gonads much enlarged and turgid, distinctly granular; wall very compact and easily broken; ova entirely opaque without translucent peripheral ring; cells not totally rounded; from 0.56 to 0.65 mm in diameter.

Stage V. Mature; gonads similar to Stage IV; ova totally rounded, whose diameter ranged from 0.66 to 0.95 mm.

Stage VI. Mature; ovaries enlarged; eggs translucent and easily dislodged from the follicle or loose in the lumen of the ovary; eggs may be set free by external pressure; size varies from 0.96 to 1.25 mm.

Stage VII. Half spent; ovarian walls loose; all eggs glassy transparent and free in the lumen.

Stage VIII. Spent; ovaries transparent, enlarged but hollow and flaccid, walls very loose, often folded; unspawned ova either free in the lumen or in the folds of the ovary; ova similar to Stage II.

The table of eight maturity stages used in this study is based mainly on the classification agreed upon in the International Council for the Exploration of the Sea (Wood, 1930), with very slight modifications. However, as in the case of other tropical fishes, Stages VII and VIII are not observed in the samples, and only a very few fish in Stage VI were seen.

Gonads from the first three to five fish examined for sexual maturity were placed in properly labelled vials with 2 per cent formalin in preparation for the measurement of the ova diameters. This was done in the following way: a portion of the ovary was teased out and laid on a slide, then the diameters of developing eggs were measured by means of a micrometer eyepiece with a value of 0.1 mm for each micrometer unit. The eggs were not always perfectly symmetrical due to the effects of preservation. To avoid biased selection of either the longest or the shortest diameter, the micrometer eyepiece was placed horizontally and the diameter parallel to the gradation on the micrometer measured. This has been found to be the most reliable procedure and the method that gave the most satisfactory results as shown by Clark (1925) and has since been adopted for egg measurements. Clark (1934) made tests as to the effect of the preservative on the size of the egg and found no apparent shrinkage or swelling. Clark (op. cit.)

made also test measurements of the eggs from the different parts of the gonad and found no significant difference in the relative number of eggs in each size group. June (1953) and Yuen (1955) have shown also that there is no difference in ova-diameter frequencies in different portions of the same ovary. Thus, in the present work, no selection of region was done. Measurements were made on not less than 200 eggs from each fish. A total of 60 fish were measured for the diameters of their eggs.

Fecundity.—Fecundity study was done mainly by counting the number of eggs. Only preserved specimens of Stages IV to VI were used for this purpose. Since actual counting of all eggs is considered impractical, it being tedious and time consuming, a modified volumetric-displacement method was employed. This method was originated by Taft [Shapovalov and Taft (1954)]. Both ovaries of each fish were placed in a vial with 2 per cent formalin and kept there until the time the eggs separated from one another by vigorous shaking of the bottle. Furthermore, a low power binocular microscopic examination of the ovary was done to make sure that no more eggs remained adhering to the walls of the ovarian tissue after each shaking. When all eggs have been separated from one another, the settling or displacement volume of the eggs was measured in a graduated centrifuge tube. Then the vial was shaken and while the eggs were being agitated, 1 cc of the known volume of egg suspension was withdrawn by an improvised pipette and the eggs placed to a Sedgewick-Rafter Counting Chamber, which is 1 cc in volume. The counting chamber was covered by a glass slide which was divided into squares. The procedure for counting the eggs was to start from the first square at the upper left hand corner of the slide, working down the first row, up the second, down the third and so on to the last square. To minimize errors, three-trial counts of every egg sample were made. The average of the three counts represented the total number of eggs per female fish. The total number of eggs [3] was calculated as follows:

$$\text{Number of eggs} = \frac{\text{Total volume of eggs} \times \text{number of eggs counted}}{\text{Volume of egg suspension counted}}$$

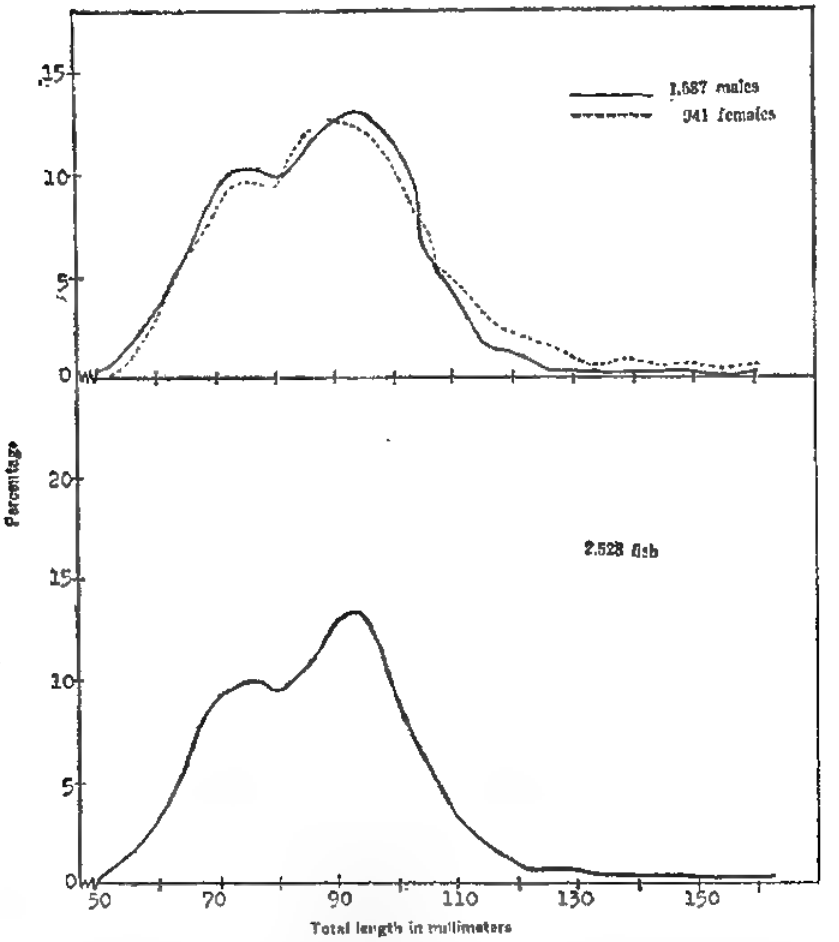
[3] Tiews, Klausdius. Lecture notes prepared for the International Training Center on the Methodology and Techniques of Research on Mackerel, Rastrelliger. Bangkok, Thailand, FAO, Rome 59/2/1404 (1958). M.S.

RESULTS AND DISCUSSION

Length-frequency method of age determination.—The length-frequency method for obtaining estimates of age of fish has found application among fishery biologist ever since Petersen first used it in 1892 in his work on *Zoarces viviparus* [Yapchiongco (1949)]. It involves the study of the progression of modes in length-frequency distributions according to time. Peterson (op. cit.) showed that the fish lengths fluctuated around certain modes. He believed that each major mode or peak represented a year group. Many investigators have successfully applied this method and found it quite accurate when checked against annular rings on scales, otolith, or vertebræ. Tropical fishes, however, because of fairly constant rate of growth the year round, do not exhibit similar ring markings on the scales, etc. Hence the only possible way of determining age is by use of the length-frequency method, which at best is still approximation.

By inspection of the length-frequency curves (Figs. 4a and 4b) two distinct modal groups can be recognized. There modes indicate the number of age groups in the sample. The first includes fish ranging in length from 65 to 80 mm with a mode at 75 mm while the second group ranges from 85 to 100 mm. This condition is true for both sexes. However, while the second in females falls on 90 mm, the corresponding mode in males is at 95 mm. This may indicate that in this particular fish the females mature a bit earlier than the males of the same age group. It is most probable that other age groups are present as shown by the few big fish in the sample. The disappearance of the larger fish from the sample indicate the effect of the fishery upon the fish population. However, the rapid growth rate of the younger age group results in the overlap of age groups and as such, the larger fish of the younger age group cannot be distinguished from the smaller fish of the next older group.

By examining Figure 1, we may be able to determine by the change in the dominant size groups from month to month, the growth rate of the fish. Fish which entered the fishery in December, 1956 with a size of 77 mm remained in the fishery until April, 1957 showing an average size of 97 mm after which the age group disappeared. This shows an average length increase of 20 mm in four months. In January, another age group at 72 mm was the most dominant in the fishery. This



Figs. 4a, 4b. Length frequency polygons of the theraponids; sexes separate in upper graph, sexes combined in lower graph.

persisted up to July, forming the most dominant group available in May, June and July at an average size of 97 mm. It may be noted that the increase in size is 25 mm in 5 months. Another age group that entered the fishery in May at 62 mm became the most dominant catch from August at 77 mm to September at 83 mm. In 4 months the increase in length is 21 mm.

From the data and in the absence of definite structures to determine the growth rate of the fish, there is sufficient evidence to indicate an increase in length of about 25 mm for a period of five months when the fish became available to the fishery or a growth rate of approximately 5 mm in one month.

Length-weight relationship.—The "Cube Law" of Spenser [Keys (1928)] was an attempt to express the rate of growth in fish. Since then the Law has been repeatedly used to measure rate of growth through a length-weight relationship. Essentially, the Law is: if, as the fish grows, it does not change its form or density, the weight of the fish will be proportional to the cube of its linear dimension. The equation for the Law is $W = kL^3$, where W is the weight of the fish, L is length, and k, a constant. For some species the formula has been found true as was shown by Beverton and Holt [Chatwin (1957)]. However, for other species with increasing age, changes in the morphology of the fish take place so that a higher or lower exponent other than the cube has to be used. For instance, in the California sardine it is 2.9353 [Clark (1928)], in the California barracuda, 2.983 [Walford (1932)], in the dover sole, 2.97 [Hagerman (1952)], and in the brook trout, 3.11061 [Allen (1956)].

In this study the modified Cube Law was applied using the exponential equation $W = aL^n$ in its logarithmic form, that is, $\log W = \log a + n. \log L$. The length-weight curve (Figure 5) for the 247 females was fitted from the following equation:

$$W = 0.0002238\ L^{3.2390730}$$
$$\log W = -5.3499109 + 3.2390740\ (\log L)$$

The equation for the 257 males is:

$$W = 0.0006156\ L^{3.1674998}$$
$$\log W = -5.210721 + 3.1674998\ (\log L)$$

The weight of the males tends to increase at a slightly lesser rate in relation to the length as shown by the difference in the values of n (Figure 6). In females, it is 3.2390740 while in males it is 3.1674998, giving a difference of 0.0716. With both sexes combined (Figure 7) the equation is:

$$W = 0.0004597\ L^{3.230442}$$
$$\log W = -5.337521 + 3.230442\ (\log L)$$

The goodness of fit of the curve as tested by the Chi-Square test shows that there is no significant difference between the actual average weight and the theoretical or expected average in either sex.

Spawning season.—Examinations of a large number of females in a one-year period of investigation showed that the silvery theraponid fish spawns every month throughout the year as shown by the presence of mature fish in the monthly

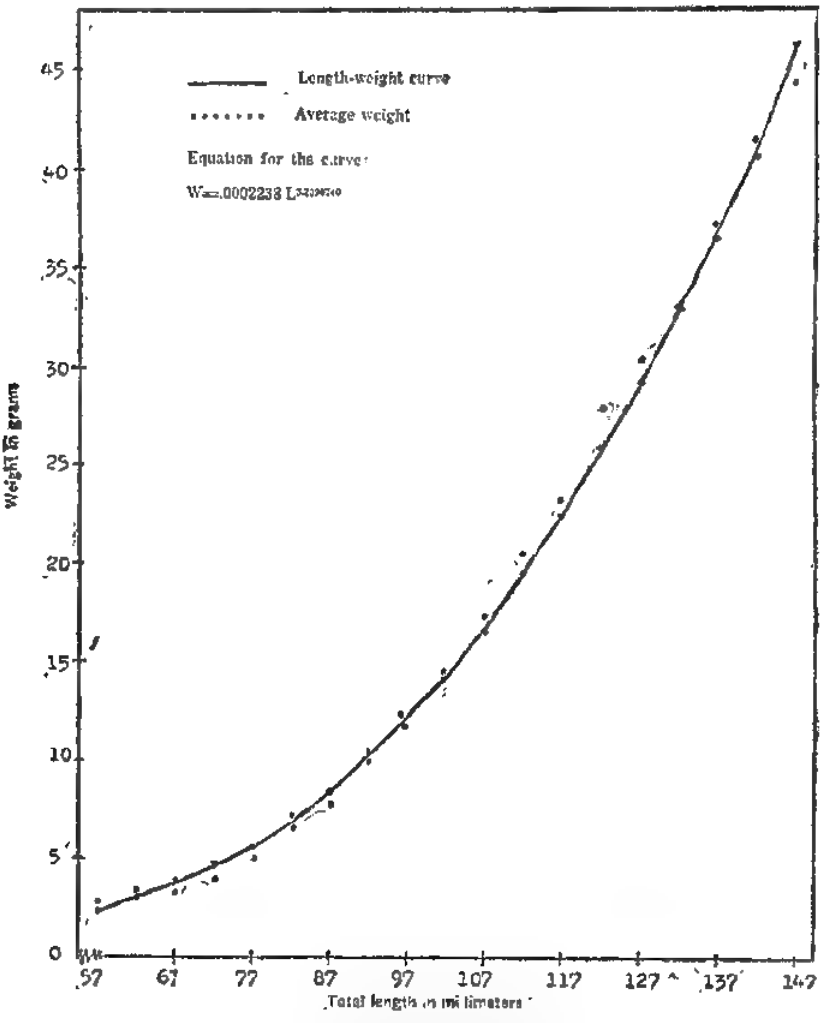


FIG. 5. Length-weight curve of 247 females silvery theraponid calculated by the method of least squares.

samples (Table 4). Figure 8 gives a summary of the spawning season as indicated by the percentage relation between spawning females and all females. It was observed, however, that spawning activity varies from month to month, with December as the month of least activity. This is shown by spawners as constituting only 3.3 per cent of all females under study. With slight fluctuation, the percentage of mature fish increased from January to March only to wane again progressively towards the month of June. Increased spawning activity was resumed

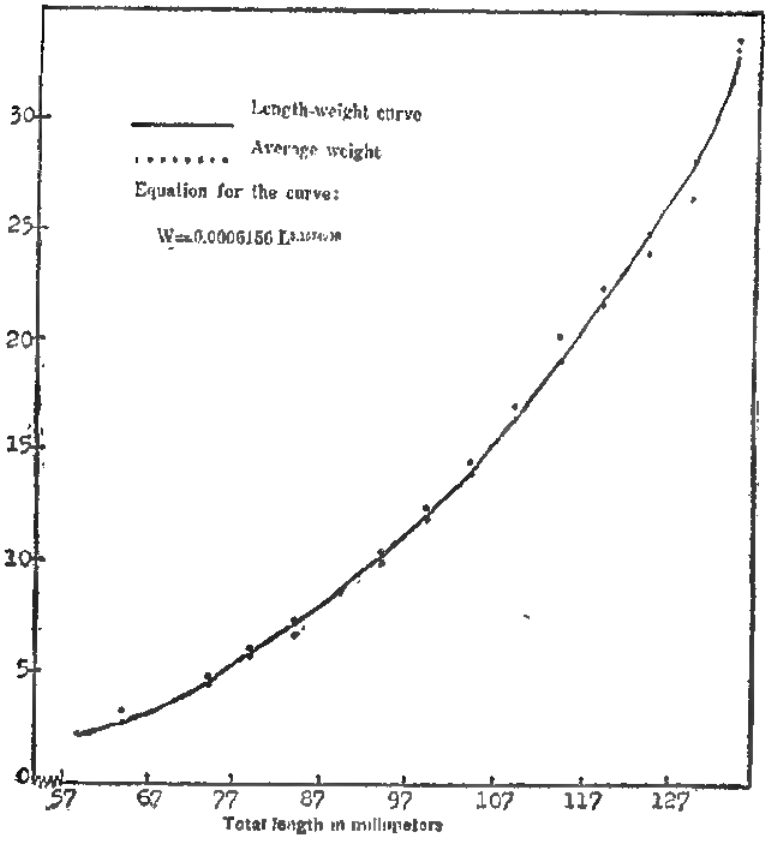


FIG. 6. Length-weight curve of 257 male silvery theraponid calculated by the method of least squares.

in July through August and culminating in September as the peak month with 64.8 per cent of the month's females in spawning condition, followed by a lull in October and November. The varying percentages of mature fish every month seem to indicate that the spawning groups in the population spawn every month. The possibility, however, of the same group spawning every month was not ascertained. There is no evidence to indicate periodicity in the spawning time, for the samples showed that while in some fishes the eggs are just starting to mature, other females were found to have maturing groups at various intermediate stages. The growth of the eggs was traced from their first origin from the immature (Stage I) and maturing (Stages II to IV) class to the mature size (Stage V) by the

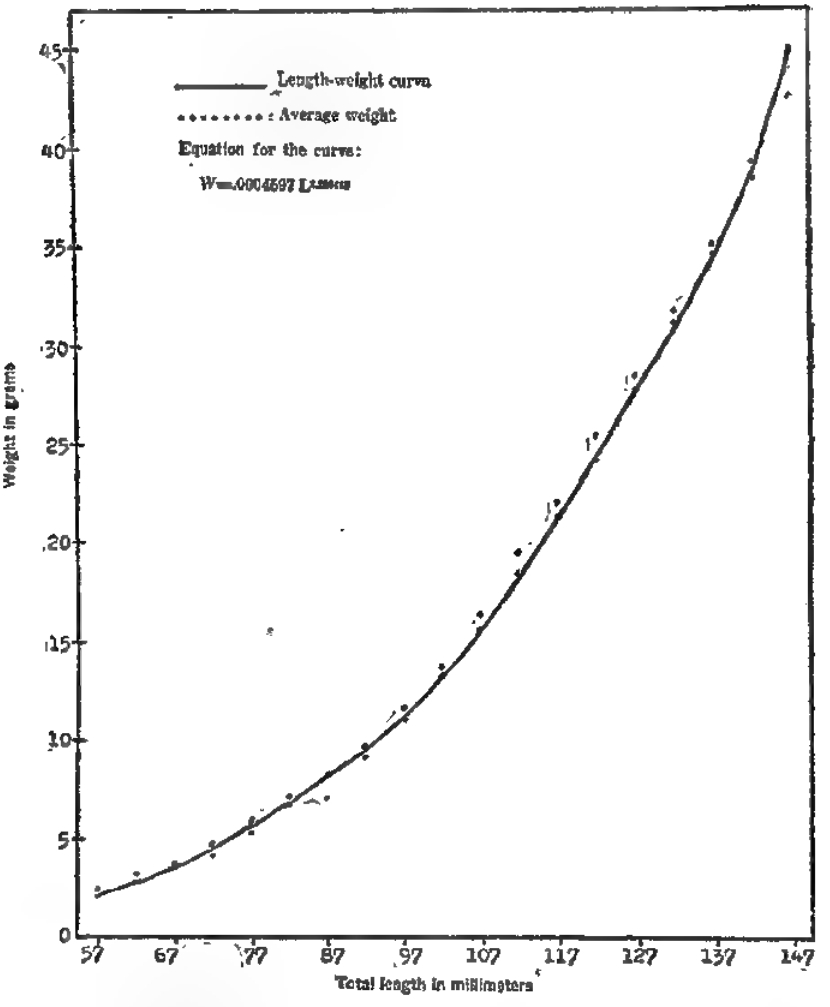


FIG. 7. Length-weight curve of 504 males and females theraponid calculated by the method of least squares.

measurement of ova-diameter, given under methods. Figure 9 shows the frequency of the first six stages taken from the collection.

Another evidence that points to the protracted spawning activity of the fish is the simultaneous occurrence of maturing and mature eggs in mature females that were examined monthly. It will also be noted from Table 4 that except for the month of September, the majority of fish sampled were maturing. In September, the majority of the fish sampled were matured.

TABLE 4.—*Table on sexual maturity.*

Total length	December, 1956			January, 1957			February			March			April			May			June			July			August			September			October			November 1957			Total			Total number of fish per group length	Percentage			Percentage	
	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g	Mat.	Im.	Mat'g.	Mat	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.	Im.	Mat'g.	Mat.		Im.	Mat'g.	Mat.	Im.	Mat
50– 54													1 (.7)			2 (3.3)																			3		3		100.0			100
55– 59		2 (2.2)			3 (13.0)						2 (1.4)		4 (3.0)	2 (1.5)		4 (6.7)	2 (3.3)		3 (5.7)			3 (3.3)			2 (5.7)			6 (4.8)	3 (2.4)			1 (1.6)	1 (1.0)			1	14	5	20	5.0	70.0	25.0	5	95
60– 64		3 (3.0)			1 (4.4)	1 (4.4)					3 (2.1)		4 (3.0)	3 (2.3)		6 (10.0)	1 (1.6)		2 (3.8)			5 (5.6)			5 (14.3)			7 (5.6)	7 (5.6)		3 (3.0)	2 (2.0)		1 (1.0)	2 (2.0)		55	12	67	82.1	17.9		100
65– 69	1 (1.1)	14 (20.0)			1 (4.4)	2 (8.7)		6 (16.4)	2 (5.1)		2 (1.4)	1 (.7)	6 (3.0)	4 (2.3)		3 (5.0)	1 (1.6)		1 (1.8)			3 (3.3)			3 (8.6)			5 (7.2)	2 (3.2)		2 (3.3)	2 (3.3)	1 (1.0)	3 (3.0)	1 (1.0)	2	54	13	69	2.9	78.3	18.3	2.9	97.
70– 74		9 (10.0)			1 (4.4)			6 (16.4)			5 (3.6)		6 (1.6)	4 (3.3)		4 (6.7)			1 (2.2)			4 (4.3)			1 (2.9)			4 (6.4)			2 (3.3)	2 (3.3)	1 (1.0)	3 (3.0)		1	29	25	85	1.2	69.4	29.4	1.2	98.
75– 79	1 (1.1)	14 (15.4)	1 (1.1)		4 (4.4)			3 (7.7)	2 (5.1)		4 (2.8)		4 (3.0)	3 (2.3)		3 (5.0)	4 (6.7)		6 (13.3)			4 (6.7)			5 (14.3)	1 (2.9)		10 (8.0)	11 (8.8)		3 (3.3)	3 (3.3)		3 (3.0)	2 (2.0)		67	27	94		71.3	28.7		100.
80– 84	1 (1.1)	9 (9.9)	1 (1.1)		1 (4.4)			7 (17.9)			4 (2.8)		4 (3.0)	3 (2.3)		6 (10.0)	1 (1.6)		6 (13.3)			3 (5.0)			5 (14.3)	2 (5.7)		5 (4.0)	6 (4.8)		3 (3.3)	3 (3.3)		1 (1.0)	2 (2.0)		89	40	129		69.0	31.0		100.
85– 89		10 (12.2)			1 (4.4)		1 (2.6)	6 (17.4)			11 (7.8)		10 (3.0)	6 (4.0)		7 (11.7)			8 (22.2)	1 (2.8)		6 (6.4)	1 (2.9)		1 (2.9)			4 (3.2)	10 (7.9)		5 (5.7)	5 (10.0)		1 (1.0)	3 (2.0)		87	33	120		72.5	27.5		100.
90– 94		4 (4.6)						1 (2.8)			11 (7.8)		27 (20.0)	1 (.7)		6 (10.0)			1 (2.8)			3 (3.2)			1 (2.9)			3 (2.4)	10 (8.0)		3 (3.6)	3 (3.6)		1 (1.0)	3 (3.0)		63	30	93		67.7	32.2		100.
95– 99		7 (7.8)						2 (5.6)			13 (9.2)		19 (14.4)	1 (.7)		1 (1.6)	1 (1.6)		3 (8.3)			2 (2.1)							7 (5.6)	1 (.8)		1 (1.6)	1 (1.6)		32	24	56		57.2	42.8		100.
100–104		3 (3.3)			2 (8.7)			3 (7.7)			16 (11.4)		8 (6.8)			4 (6.7)	2 (3.3)		3 (6.9)			2 (2.1)			2 (5.7)				3 (2.4)		1 (1.6)			1 (1.0)	4 (4.0)		24	4	28		85.7	14.3		100.
105–109					2 (8.7)			1 (2.6)			7 (5.0)		8 (6.8)			3 (5.0)			3 (6.7)			1 (1.6)			1 (2.9)				1 (.8)					3 (3.0)			25	4	29		85.5	14.5		100.
110–114		1 (1.1)						1 (2.6)			2 (1.4)		5 (4.2)	1 (.7)		1 (1.6)						1 (1.6)			1 (2.9)				1 (.8)					5 (5.0)	4 (4.0)		19	10	29		65.5	34.5		100.
115–119					1 (4.4)			1 (2.6)			3 (2.8)		4 (3.3)			1 (1.6)						1 (1.6)			1 (2.9)				1 (.8)					6 (6.0)			13	3	16		81.3	18.7		100.
120–124		1 (1.1)						1 (2.6)			3 (2.4)		3 (2.5)																1 (.8)					5 (6.0)			11	2	13		84.4	15.6		100.
125–129		1 (1.1)									2 (1.4)		1 (.8)			1 (1.6)			1 (2.2)										1 (.8)					5 (6.0)			5	2	7		71.5	29.5		100.
130–134		1 (1.1)						1 (2.6)			2 (1.4)		1 (.7)																					1 (1.0)	1 (1.0)		2		2		100.0			100.0
135–139		1 (1.1)									1 (.7)																										3		3		100.0			100.0
140–144		2 (2.2)																																			1		1		100.0			100.0
145–149																												1 (.8)																
150–154		1 (1.1)																																			1		1		100.0			100.0
155–159	2	85	3		19	4		36	3		90	31	107	25		46	14		43	2		49	17		27	8		44	31		31	20	4	75	22	6	657	9 0	813					
Monthly total	2.2	90	3.3		23	17.5		39	7.7		141	36.2	132	18.7		69	23.0		45	4.4		60	18.4		35	22.9		125	64.8		60	48.1	4.0	101	22.0									
Per cent		94.6			82.8			92.5			63.8		81.0			76 4			95.6			81.4			77.3			35.2			51.5			74.8									

Numbers inside parenthesis denote percentage

While November and December yielded maturing and mature fish, yet at the same time these two months are the only months of the year when immature fish occurred.

The occurrence of a protracted or a continuous spawning season is however, not unusual among tropical fresh-water fishes. Villadolid and Manacop (1934) reported a similar condition in *Gulaphallus mirabilis* Herre of Molawin Creek, Laguna. An almost identical phenomenon was observed by Manacop (1953) in *Sicyopterus extraneus* Herre of Cagayan River, Oriental Misamis. The fimbriated sardine, *Sardinella fimbriata* Cuvier and Valenciennes [Anicete and Yapchiongco (1960)] and the mudfish, *Ophicephalus striatus* Bloch (Yapchiongco and Demonteverde) were found to spawn throughout the year. In his work on the engraulids and clupeids of the Gulf of Panama, Peterson (1956) reports that *Anchovia macrolepidola* spawns the year round too, but the peak is in late spring or early summer. Davies (1956) also reports that the spawning season in the South African pilchard, *Sardinops ocellata* Pappe occurs throughout the year to a limited degree. Clark (1925) cited Fulton (1899) for having stated that the presence of maturing group of eggs is almost always associated with prolonged spawning season.

After an extensive examination of the eggs was done, some evidence show that each fish spawns more than once during the season. The egg measurements of a number of specimens revealed that immature, maturing and mature eggs are found simultaneously in a mature ovary at all times of the year. This is shown in Figure 10, which shows the frequencies of 772 egg diameter measurements from a mature (Stage VI) fish. Three groups are distinguished; namely, a great number of immature group found at the extreme left of the graph, measuring from 0 to 0.19 mm made up of totally transparent, polygonally shaped cells, resembling the nest of a honey bee; the maturing group, measuring from 0.20 to 0.65 mm in diamter and composed of eggs from Stages II to IV; and the mature group, the number of which is very slight in proportion to the great quantity of the immature eggs.

Apparently the multiple spawning habit of the silvery theraponid is not a unique feature of the fish. In *Arius* sp? [Mane (1929)], the Philippine tunas such as *Neothunnus macropterus* (Schlegel), *Katsuwonus pelamis* (Linnæus), and *Euthynnus*

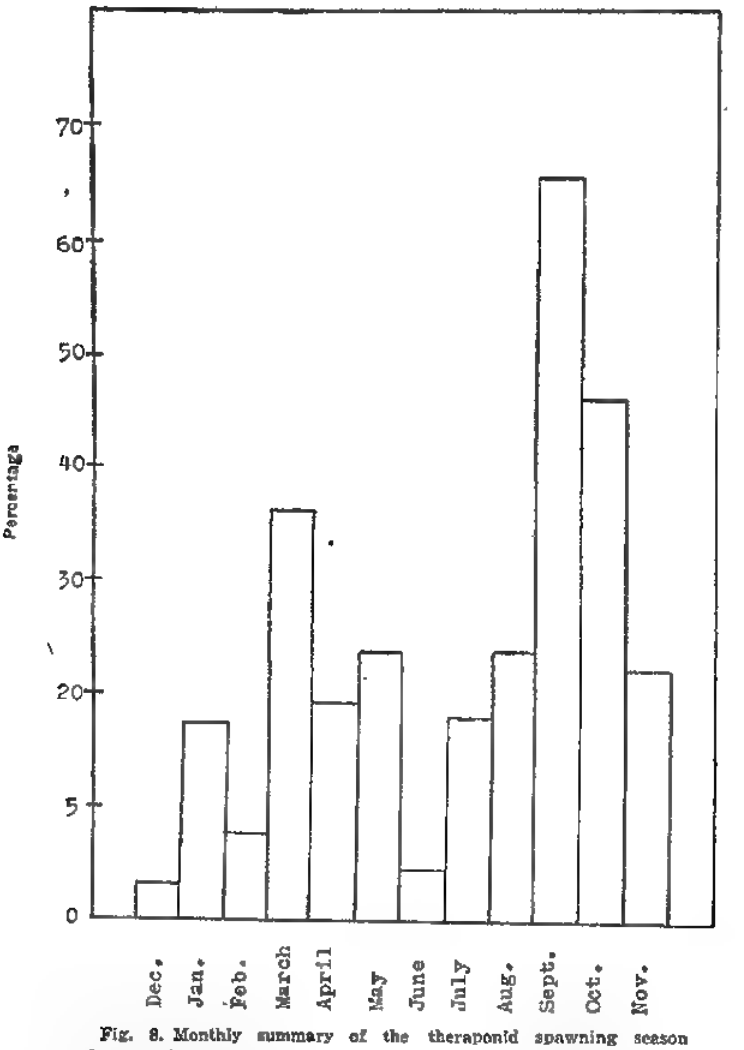


Fig. 8. Monthly summary of the theraponid spawning season showing the relationship between total female to spawning female by percentage.

yaito Kishinouye [Buñag (1905)], and *Sicyopterus extraneus* (Manacop, op. cit.) same condition holds true. Parallel phenomena have been reported in the cases of *Leuresthes tenuis* [Clark (1925)], *Atherinopsis californiensis* [Clark (1929)], *Sphyraena argenteus* [Walford (1932)], *Sardina caerulea* [Clark (1934)], *Scomber scombrus* [Sette (1943)] and *Hypomesus pretiosus* [Yapchiongco (1949)].

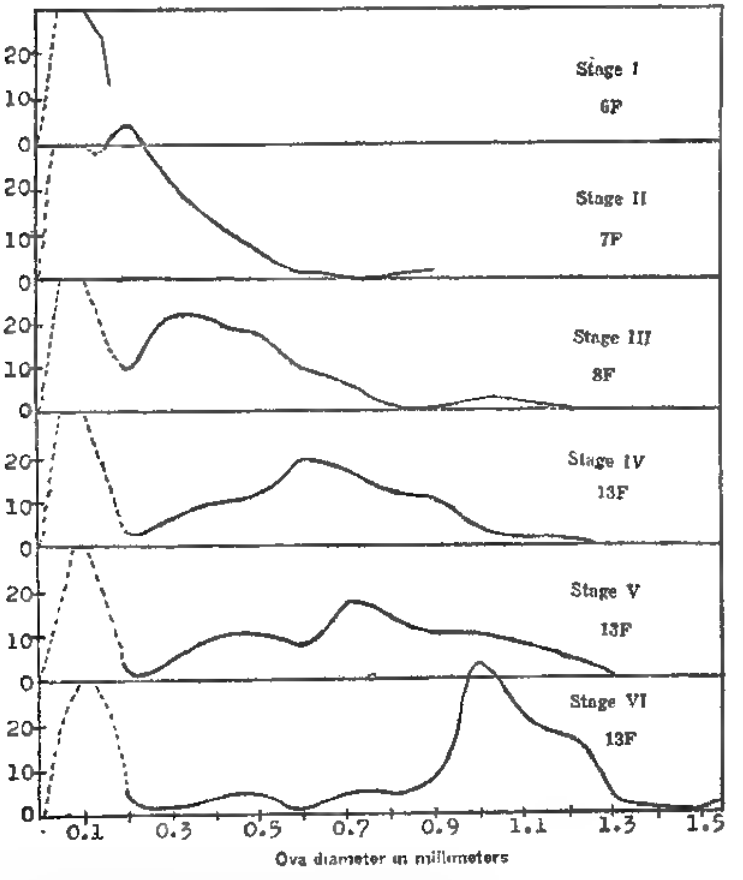


FIG. 9. Ova-diameter frequency polygons showing the growth of the eggs to maturity. Each frequency represents measurements of not less than 200 ova from one or more females. Materials taken from collection during the period of investigation.

Size at sexual maturity.—In the determination of the size at sexual maturity only the female fish were considered. The ovaries of all females were examined and the stage of maturity of the individual fish ascertained. The presence of mature or Stage V eggs indicate the maturity of the fish.

Table 4 shows the results of these determinations. Of the 913 females under review, 250 were mature, 657 maturing and 6 specimens were immature. Since the maturing group showed ovarian egg differentiation also, then the presence of both the maturing and mature eggs served as the criterion for

sexual maturity. The smallest sexually mature fish were found in the size group from 55 to 59 mm, yet it would not be right to conclude rashly that in the native haunt, all female fish within the above size group are all sexually mature since in the size groups following it, sexually immature fish were found. Fishes, however, from 85 mm up to the longest fish (156 mm) were all sexually mature.

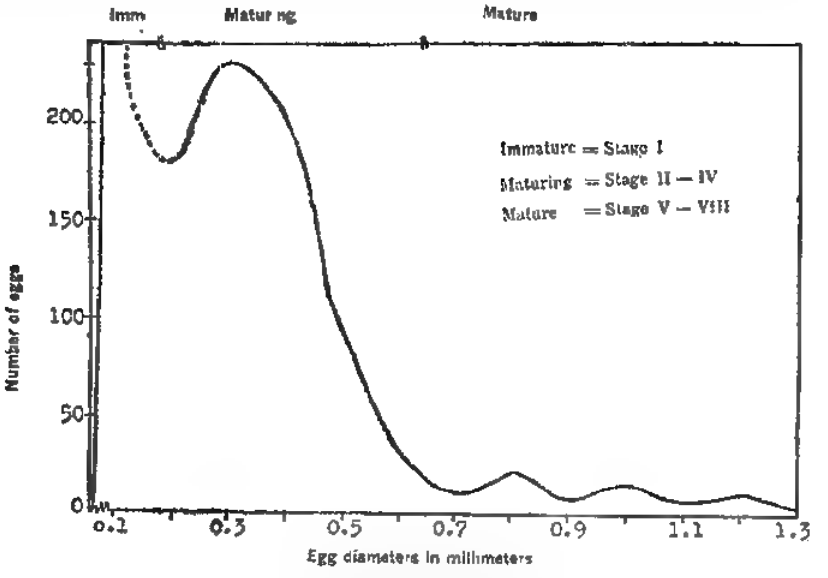


FIG. 10. Frequency polygon of the diameters of 772 ova measured from a mature theraponid 134 mm in total length.

Sex ratio.—Although the male specimens were not considered for examination in the determination of the spawning season, they were nevertheless measured for two purposes, namely, for age determination and sex ratio studies. Table 1 shows that in the monthly collection, the males predominantly outnumbered the females except in September, when the females tipped the balance in their favor by eight. The collection showed that out of a total of 2,528 specimens, only 941 were females and 1,587 were males, or a sex ratio of slightly less than 2 males to every female. This, however, is not surprising. Among fishes it is not uncommon for the males to outnumber the females, at least during the spawning period. For instance, in his work on California barracuda, Walford (op. cit.) shows that males tend to outnumber the females. In referring to Kendall's work (1926) Walford also cited the same condition to hold true with

the smelts. Scofield (1931) working on stripped base gave the average yearly sex ratio in percentage of 56.5 to 43.5 in favor of the male side. Yapchiongco (op. cit.) showed that in the surf smelt, *Hypomesus pretiosus*, the spawning population was predominantly males to the extent of almost 3 to 1 female. Conversely, there are also cases wherein the females predominate as in the grunion and jack smelts (Clark, op. cit.), or in the European plaice [Hefford (1909)]. Hagerman (1952) got a ratio of 3 females to 1 male in the dover sole, *Microstomus pacificus*, but noted that beyond 50 cm no males were caught. In the South African pilchard, *Sardinops ocellata* Davies (1956) found that the mean ratio of female is 46 to 44. This, however, varies with the size of the fish. Beyond 22.5 cm all fishes examined were females.

Fecundity. The determination of the number of eggs spawned by each fish is found rather difficult due to the multiple spawning habit of the species. However, the approximate or relative number of maturing eggs contained in the whole ovary was estimated as described under methods. The results are shown in Table 5. It includes the total length of the fish and the weight of the gonads. Only fish from Stages IV to VI were used. The egg counts are expressed to the nearest hundred.

Table 5 shows that like in most other fishes, the number of eggs increases with increasing increment of both length and weight. It also shows that in a fish 100 mm long with an ovarian weight of 1.5 grams, there are 37,500 eggs representing the maximum calculated fecundity in the present study. However, it may be noticed that in some cases, even with increased length, the weight either remains the same or decreases and consequently, the number of calculated eggs varies. This can be explained by the fact that an individual fish spawns several batches of eggs during the season, hence it is safe to say that some of the fishes under study have already spawned partially before they were collected.

The method here applied in the enumeration of the eggs is what is conveniently termed modified volumetric displacement method as given by Tiews (op. cit.).

Other workers on fecundity employed other methods. For instance, Orcutt (1950) calculated 11,000,000 in a fish 565 mm in standard length with an ovary 262 mm long by using Clark's

TABLE 5.—*Egg counts of 30 fish of varying lengths showing the weights of gonads.*

Total length	Weight of gonads	Calculated number of eggs
mm	gms	
73	0.5	8,900
81	0.4	6,700
81	0.4	6,200
83	0.9	16,700
84	1.1	12,500
85	0.8	1,800
87	0.8	24,200
89	0.9	16,900
89	0.8	23,800
92	1.0	12,500
93	0.5	12,600
93	1.5	22,600
94	1.2	37,400
94	0.5	15,900
95	0.6	17,000
96	0.3	3,400
97	1.4	17,300
98	1.4	34,900
100	1.5	37,500
100	0.7	15,900
102	0.7	3,300
103	1.0	13,400
104	1.1	10,200
104	0 6	15,900
104	1.1	16,600
104	1.1	21,600
105	0.9	25,600
107	1.2	18,400
126	2 4	29,300

method (op. cit.) which was done by counting a gram of eggs and then multiplying the number of counted eggs by the combined weight of both ovaries. This same method was used by O'Connell (1953) in his work on the life history of the cabezon, *Scorpaenichthyes marmoratus* Ayres. An actual count of the number of eggs on all specimens caught was made by Carbine (1944) in the northern pike, *Esox lucius* L. He got an average of 32,200 eggs (ranging from 7,691 to 97,273) for fish 15.7 to 35 inches in length. Inspite of the great accuracy in the actual count method, Lagler (op. cit.) recommended favorably the volumetric method as good substitute for the approximation of the number of eggs.

SUMMARY

The silvery theraponid or *ayuñgin* (local name) is found only in the Philippines, hence endemic. It abounds in all portions of Laguna de Bay and serves as food for the laboring class as a result of abundance and consequently lower prices within the reach of the masses.

The fish properly belongs to species *Therapon* (*Datnia*) *plumbeus* (Kner).

The length-frequency study shows that there are, at least, two distinct modal groups, the first, ranging from 65 to 80 mm and a second group ranging from 82 to 105 mm. The presence of other older age groups is indicated by the few large fish caught which cannot be included in the second age group. The vulnerability of the fishes of the second age group to fishing operations may explain the presence of only few big fishes in the sample.

The weight of the fish increases more than the cube of its length; arithmetically determined it is 3.2390740 for females and 3.1674998 for males; the length-weight curve fitted from the exponential formula, $W = aL^n$ expressed in its logarithmic form. Formulæ are as follows:

for females: $W = 0.0002238\ L^{3.2390740}$
for males: $W = 0.0006156\ L^{3.1674998}$

The spawning season occurs throughout the year with evidences showing individual fish spawns more than once during the year; periodicity of spawning was not determined.

The condition mentioned immediately above rendered difficult the determination of the number of eggs spawned by each fish. However, from one female 100 mm total length an egg estimate of 37,500 was found. This is shown in Table 5.

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THE PHYSICAL AND EATING QUALITIES OF LOWLAND RICE

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SIX TEXT FIGURES

Rice is the main staple of the Filipinos; it is eaten two or three times a day. Plain boiled rice[1] is the form in which it is chiefly used. We know the loss in valuable nutrients in repeated washings and in discarding water in which the rice is cooked. However, we have not arrived at a definite proportion or ratio of water to use in cooking the different varieties of rice to prevent overflowing. The procedure of calculating water by immersing the hand till the water reaches the middle node of the middle finger is still the common practice. Hence, cooked rice may be improperly cooked, either too dry or too wet and much of the vitamin content is lost by cooking in an excess of water. Vitamins present in rice before cooking can be preserved by using the correct amount of cooking water to prevent overflowing thus benefiting the consumer.

The rice varieties under study which came from the Bureau of Plant Industry are: Raminad; Tjahaja, S. Kechil, Peta, and B.E.–3. These are lowland varieties which are recommended by the Philippine Seed Board and classified according to grain type as follows:

Raminad, Tjahaja ..... long grain varieties
Peta, B.E.–3 ..... medium grain varieties
S. Kechil ..... short grain variety

The objectives of this study are: (*a*) to determine some physical qualities of rice such as water uptake ratio and volume of cooked rice, and (*b*) to determine the factors affecting palatability of rice by evaluating the following qualities: color, cohesiveness, off-flavor, and doneness.

[1] Plain boiled rice—rice is boiled with a measured amount of water (usually 1½ times the amount of rice) until all the water is absorbed. Cooking is continued over low heat until rice is done.(12)

Several studies on the physical and eating qualities of rice that have been conducted locally and abroad were used as bases for this study. In determining the physical qualities of rice, Batcher, et al.(1) calculated the water uptake ratio of cooked rice as the weight of cooked rice divided by the weight of raw rice. Planck(8) and Joseph and Planck(8) in their studies found that the variety of rice affects rate and extent of absorption of water. Dufournet and Rakotomanana(4) observed that increase in weight and volume are varietal characteristics and are also directly proportional to the time of boiling in water within the permitted range. Odor and taste are affected by the length of cooking.

The cooking quality of the different types of rice (long, medium, and short grain) have been studied by Irwin.(6) In these tests, the taste panel found that the cooking quality of rice is dependent on, or correlated to its variety. Jenkins(7) in his studies showed that medium grain type varieties tend to be slightly sticky or moist when cooked. The long grain varieties are easier to cook. Ferrel, et al.(5) found that short grain or pearl rice are quite sticky when cooked. Cada, et al.(3) conducted an experiment on the culinary qualities of the different varieties of rice from Indonesia. Results of their experiment showed that (long grain type) was considered very good in eating quality based on appearance, texture, stickiness of the grains and palatability of the cooked rice. Studies of Oñate and del Mundo(10) on upland rice at the College of Agriculture showed that varietal differences influenced the eating quality of rice.

EXPERIMENTAL PROCEDURE

Physical test.—Four hundred (400) grams sample of rice was washed twice with 550 grams water. The water was decanted and the washed rice was drained for five minutes in a strainer. The first and second washings were weighed separately and the weights were recorded to determine the amount of water absorbed in the washings.

To determine the best proportion of water, three levels of water were tried; namely, 550 grams, 600 grams, and 660 grams. To minimize variables in cooking, rice was cooked the same hour from 9:00 to 11:00 o'clock in the morning for all trials using four uniform pots on the top burner of a range. The

cooking time ranged from 23 to 30 minutes (23, 25, 28, and 30 minutes) for each level of water used.

Water uptake ratio was calculated as the weight of cooked rice divided by the weight of raw rice.(1) Volume of cooked rice was calculated by the formula $V = \frac{1}{3} h [B + B' + \sqrt{B B'}]$ where h is height of cooked rice, B is top area and B' is bottom area. The volume of a frustum of a circular cone equals the sum of its bases and the mean proportional between them multiplied by $\frac{1}{3}$ of its altitude.

The trial was repeated after six months and a third trial done after one year to determine the effect of storage upon the physical qualities of rice.

Palatability test.—Four hundred grams samples of each rice variety which were washed and cooked in the same manner as for the physical test using only one level of cooking water, were cooked for different lengths of time and presented to a taste panel of twelve members at each evaluation session. The samples were rated on color, cohesiveness, off flavor, and doneness using the score-card shown in Table 1.

TABLE 1.—*Score card for palatability.*

Qualities	Sample			
	A	B	C	D
Color:				
9 white				
7 cream				
5 yellow				
3 tan				
1 brown				
Cohesiveness:				
9 sticky				
7 partially separated				
5 very sticky				
3 well separated				
1 pasty				
Off-flavor:				
9 none				
7 perceptible				
5 slightly strong				
3 moderately strong				
1 very strong				
Doneness:				
9 done				
7 slightly underdone				
5 slightly overdone				
3 underdone				
1 overdone				

To control and eliminate variables, a special room for taste panel was used. A long table was divided into six individual booths partitioned by plywood structure painted bluish-gray which was lighted from the ceiling with fluorescent lights. The

table setting in each booth was identical. Further details on the procedure used has been reported in a separate paper.(2)

The panel members were selected from employees of the Department of Foreign Affairs, Department of Justice and the Food and Nutrition Research Center who had previously passed the elimination tests for judges using Tukeys' procedure of statistical analysis.

Three trials were performed for each variety and mean scores for the different qualities of cooked rice for the different lengths of time were evaluated using a modified score card of Batcher, et al.(1) to conform with the pattern of our eating habits. A pretest of the score card was conducted by panel members which consisted of employees and trainees from the Food and Nutrition Research Center and four students. Rice samples were presented to them to determine their ratings of the qualities under study.

RESULTS AND DISCUSSION

PHYSICAL QUALITIES OF COOKED RICE

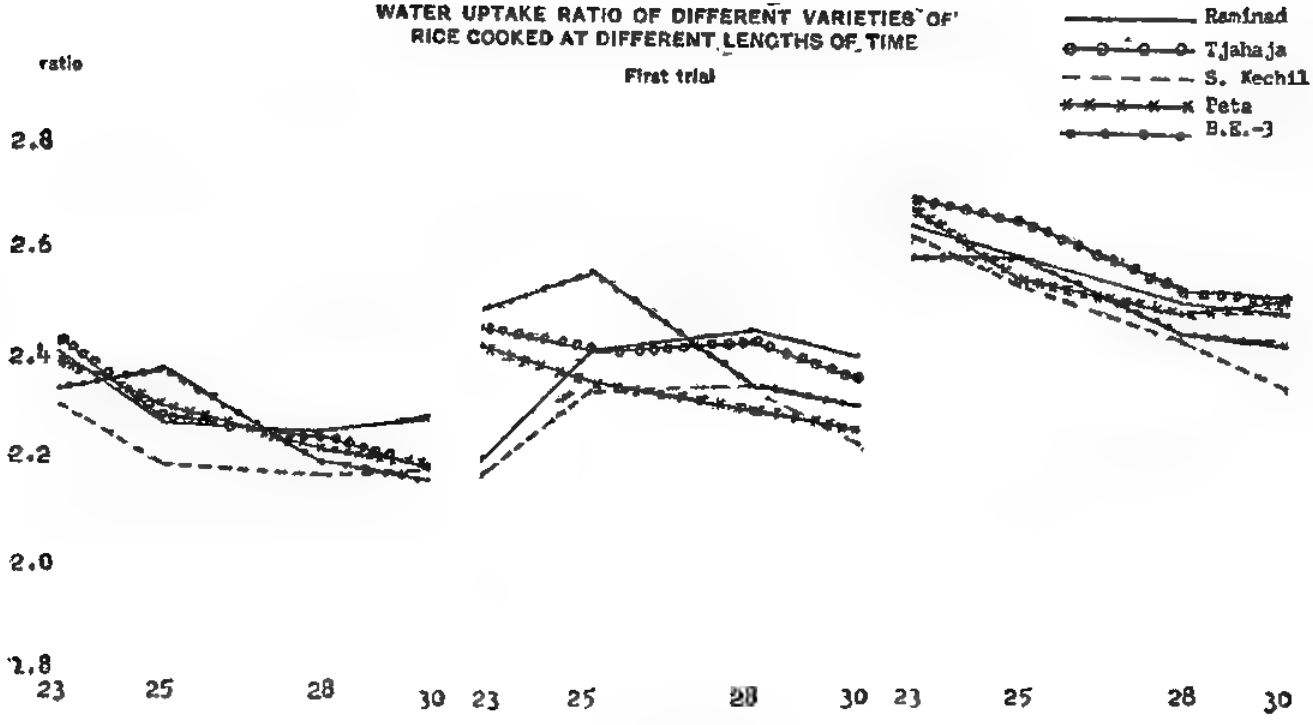
Water uptake ratio.—The general tendency in water uptake ratio was that it was lowest for all varieties with the least amount of cooking water used (Figure 1 and Table 2). As the amount of cooking time increased, water uptake ratio decreased in all levels of cooking water, although there were exceptions.

At the first trial, Raminad and Tjahaja had slightly higher water uptake ratio than the other varieties and S. Kechil, the lowest water uptake ratio. This is in line with the findings of Batcher, et al.(1) that long grain rice tends to absorb more water than either the medium or short grain variety. B.E.-3 showed the characteristics of long grain variety at 550 and 600 grams water level, but showed the characteristics of short grain variety at 660 grams water level.

In the second trial after six months, it was observed that water uptake ratio was slightly less than the first trial presumably because of loss of moisture during storage of rice.

In the third trial a year after, S. Kechil showed a higher water uptake ratio at 550 and 600 grams water level than the other varieties tested. Similar to the second trial, water uptake ratio in the third trial was slightly less than the first trial.

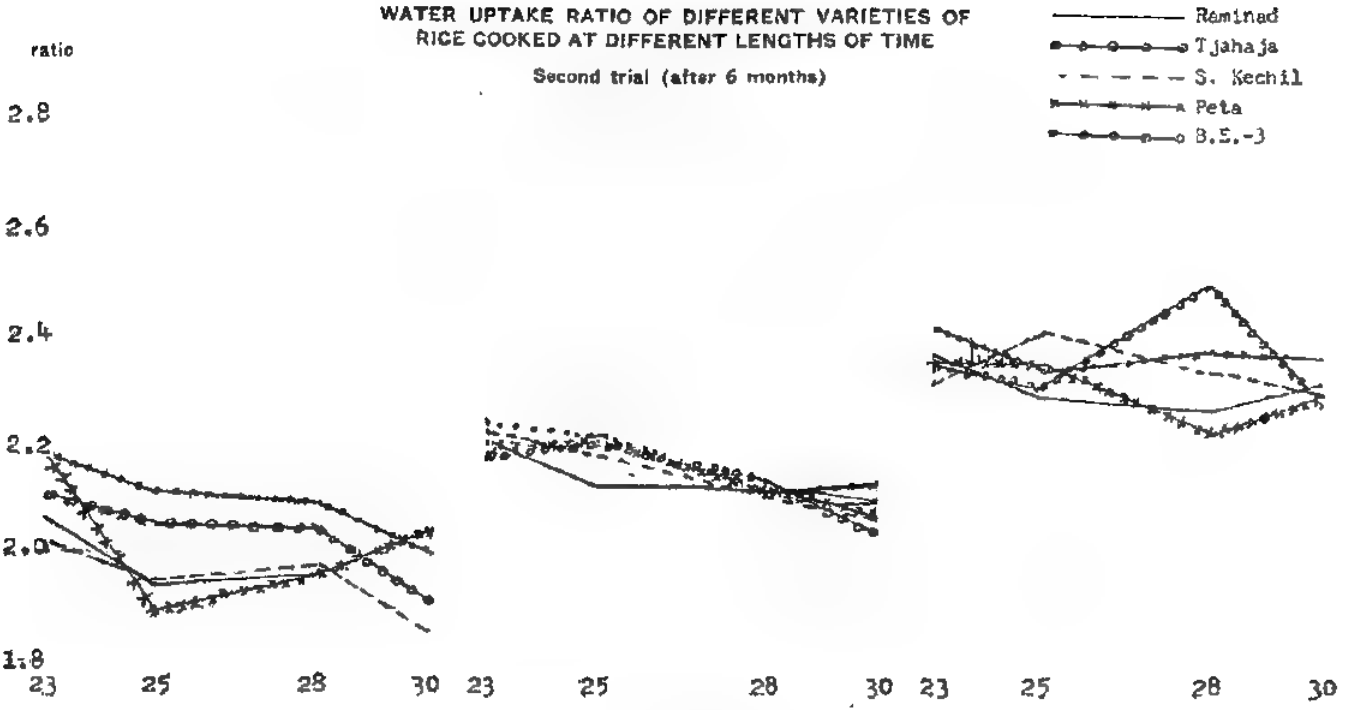
Volume of cooked rice.—The volume of cooked rice increases with increasing amount of cooking water and decreases as the



550 gms water
Fig. 1a

600 gms water
Fig. 1b

660 gms water
Fig. 1c



550 gms water

Fig. 2a

600 gms water

Fig. 2b

660 gms water

Fig. 2c

(22784)

550 gms water

Fig. 3a

600 gms water

Fig. 3b

660 gms water

Fig. 3c

TABLE 2.—*Water uptake ratio of different varieties of rice cooked at different length of time.*

Amount of cooking water used (gm)	Cooking time in minutes	Raminad			Tjahaja			S. Kechil			Peta			B. E.-3		
		Number of trials			Number of trials			Number of trials			Number of trials			Number of trials		
		first	second	third	first	second	third	first	second	third	first	second	third	first	second	third
550	23	2.40	2.07	2.28	2.43	2.10	2.22	2.30	2.01	2.31	2.39	2.17	2.25	2.33	2.18	2.25
	25	2.27	1.91	2.28	2.28	2.05	2.29	2.19	1.95	2.30	2.30	1.89	2.26	2.37	2.11	2.28
	28	2.26	1.96	2.12	2.25	2.04	2.09	2.18	1.98	2.26	2.23	1.96	2.18	2.20	2.09	2.19
	30	2.29	---	2.19	2.19	1.91	2.17	2.19	1.86	2.30	2.20	2.04	2.13	2.17	2.00	2.14
600	23	2.21	2.21	2.39	2.46	2.18	2.36	2.18	2.23	2.43	2.43	2.20	2.37	2.49	2.24	2.46
	25	2.42	2.13	2.37	2.42	2.20	2.28	2.34	2.18	2.40	2.36	2.21	2.40	2.57	2.22	2.31
	28	2.46	2.13	2.16	2.44	2.14	2.26	2.35	2.11	2.32	2.31	2.12	2.31	2.35	2.12	2.22
	30	2.41	2.10	2.24	2.37	2.04	2.20	2.25	2.07	2.38	2.27	2.07	2.18	2.32	2.13	2.27
660	23	2.66	2.37	2.57	2.71	2.35	2.56	2.64	2.32	2.57	2.69	2.36	2.48	2.60	2.42	2.60
	25	2.60	2.29	2.52	2.67	2.31	2.49	2.55	2.41	2.53	2.56	2.35	2.52	2.60	2.34	2.48
	28	2.52	2.27	2.41	2.54	2.50	2.51	2.45	2.34	2.44	2.50	2.23	2.45	2.46	2.33	2.45
	30	2.50	2.32	2.32	2.53	2.28	2.54	2.36	2.30	2.36	2.32	2.30	2.35	2.44	2.37	2.40

length of cooking time increases, following the same trend as the water uptake ratio.

Varietal differences affect the volume of cooked rice. In the first trial, Raminad and Tjahaja had a bigger volume than other varieties. B.E.–3 showed the characteristics of long grain variety at 550 and 600 grams water level. (Figure 4 and Table 3.)

In the second trial six months later, Peta and B.E.–3 had a bigger volume than other varieties as shown in Figure 5. The length of storage affects volume of cooked rice. S. Kechil which had the least volume during the first trial expanded more than other varieties after one year (Figure 6). The long grain varieties which had a greater volume in the first trial showed the least volume at the third trial. Medium grain varieties were intermediate between long and short grain types.

Water absorbed in rice washings.—Tests showed that the amount of water absorbed in washing has no significant effect on cooking quality of rice. However, water absorption during cooking affects cooking quality of rice. Rao, et al.(11) called this the swelling number or amount of water imbibed by 100 grams rice when cooked in excess water at 98°C under standard conditions. The "swelling number" was then used as an index of cooking quality.

PALATABILITY TEST

Results of the palatability tests are summarized in Table 4.

Color.—Raminad obtained the highest mean scores and was rated by the panel members as more white and less creamy than the other varieties. Differences of rating in color between the different varieties were not statistically significant at the 5 per cent level. However, at 23 minutes cooking time Raminad showed significantly higher rating than both Tjahaja and B.E.–3.

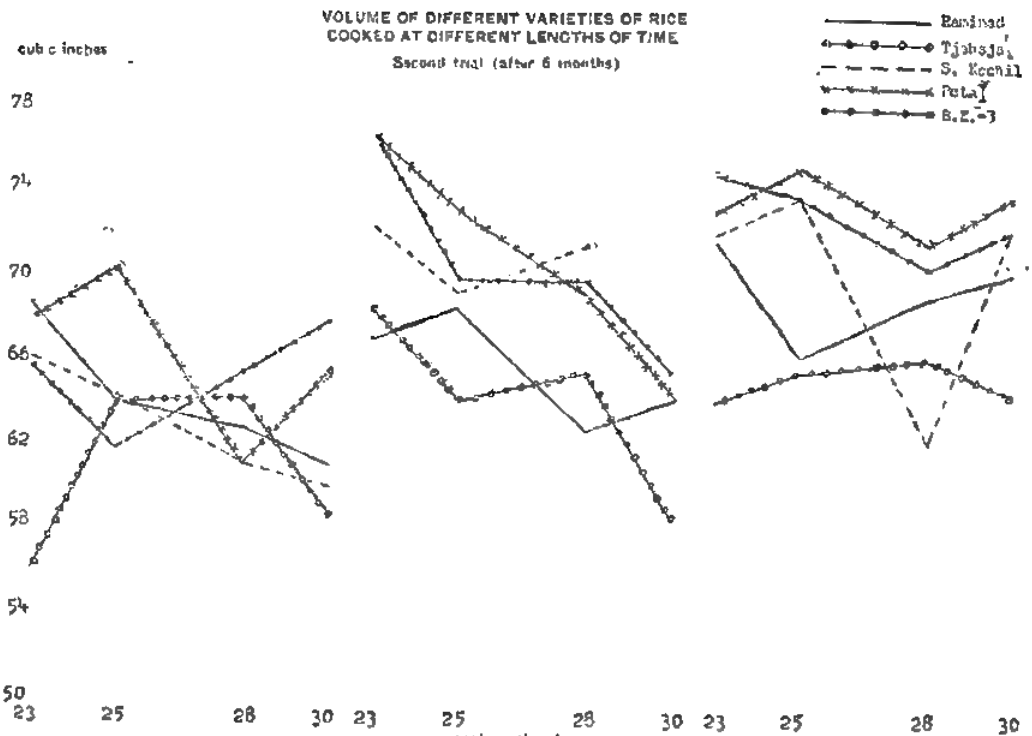
Cohesiveness.—Panel members preferred a sticky product to the partially separated cooked grains. Raminad and B.E.–3 obtained higher scores as compared with the other varieties which were significant. Although at 23 minutes cooking time Tjahaja was rated higher than both Raminad and B.E.–3.

Off-Flavor—Some off-flavors were noted in all the varieties of rice tested. It ranged from "none" to "slightly strong" and was noticeable at 30 minutes cooking time. Raminad was rated as having the least off-flavor among the varieties tested

Fig. 4a

Fig. 4b

Fig. 4c



330 gms water
Fig. 4a

660 gms water
Fig. 5b

660 gms water
Fig. 5c

VOLUME OF DIFFERENT VARIETIES OF RICE COOKED AT DIFFERENT LENGTHS OF TIME

Third trial (after 1 year)

550 gms water

Fig. 6a

600 gms water

Fig. 6b

660 gms water

Fig. 6c

TABLE 3.—*Volume of different varieties of rice cooked at different lengths of time.*

Amount of cooking water used	Cooking time in minutes	Raminad			Tjajaha			S. Kechil			Peta			B. E. 3		
		Number of trials			Number of trials			Number of trials			Number of trials			Number of trials		
		first	second	third	first	second	third	first	second	third	first	second	third	first	second	third
gm	23	64.16	68.42	65.57	66.29	55.73	69.85	65.94	56.94	74.52	69.14	67 71	74 52	69.85	65.57	69.14
	25	63.81	63.81	66.29	65.94	63.81	68.42	65.22	64.16	73.07	68.10	70.21	70.21	70 21	61.69	71 64
500	28	65.57	62.75	67.00	65.94	64.16	63 10	64.51	60.99	74.52	64.51	60.99	68 42	65.22	65.57	71.28
	30	65.57	60.99	67.71	70.56	58.53	72 00	64.16	59.93	71.28	64.51	65.57	68.78	67.35	67.71	67.71
	23	72.36	67.00	71.64	72.00	68.42	69.14	70.92	72.36	73.44	69.85	76.69	73 07	72 00	76.69	74.52
600	25	70 92	68.42	73.44	73.44	64.16	69.85	73.07	69.14	76.96	68.42	73 07	73 80	75 23	69.85	74.52
	28	70.21	62.75	65.57	70.56	65 57	65.57	70.56	71.64	72.36	65.94	69.14	72 86	70 21	69.85	69.85
	30	70.56	64.16	65.22	70.21	58.53	64 51	68.42	.....	73.86	66.22	64.51	68.42	73.80	65.57	65.57
	23	76.96	71.64	73.07	76.69	64.16	73.07	77.77	72.00	77.40	76.69	73.07	73.80	75.23	74.88	77.77
660	25	75.23	66.29	73.56	76.69	66.57	72.00	73.44	73 80	79.58	73.44	75.23	75 23	74 88	73.80	76.69
	28	72.36	69 14	70.92	74 52	66.29	74.52	73.07	62.30	73.80	71.64	71.64	74 52	73.44	70.56	76 60
	30	71.64	70.21	67.35	73.44	64.51	73.07	68.42	72.36	73.07	73.80	73.80	70.56	70.92	72.36	74.88

TABLE 4.—*Mean scores for different qualities of cooked rice at different lengths of time.*

Qualities and varieties	Cooking time in minutes			
	23	25	28	30
Color:				
Raminad	8.89±.01*	8.67±.05	8.72±.18	8.56±.18
Tjahaja	8.50±.19	8.78±.17	8.67±.09	8.00±.28
S. Kechil	8.33±.18	7.95±.20	8.00±.19	6.83±.37
Peta	7.78±.32	7.61±.39	8.28±.31	8.06±.29
B. E.-3	8.11±.02	8.06±.20	8.00±.22	7.79±.36
Cohesiveness:				
Raminad	7.11±.01	7.56±.04	7.50±.37	6.89±.41
Tjahaja	5.83±.35	6.06±.33	6.00±.36	5.22±.30
S. Kechil	5.67±.35	5.28±.30	5.78±.34	5.50±.33
Peta	5.72±.35	5.72±.36	5.94±.39	5.33±.37
B. E.-3	6.50±.20	7.11±.04	7.33±.05	7.42±.05
Off-flavor:				
Raminad	7.83±.23	8.00±.22	7.72±.29	7.22±.25
Tjahaja	6.33±.38	7.00±.35	7.22±.04	6.33±.33
S. Kechil	6.50±.37	6.28±.37	6.72±.31	4.61±.36
Peta	6.61±.44	7.00±.35	7.22±.31	6.33±.33
B. E.-3	7.39±.32	7.50±.23	7.06±.32	6.33±.42
Doneness:				
Raminad	8.65±.01	8.89±.01	8.33±.28	6.94±.46
Tjahaja	7.78±.30	7.83±.26	7.72±.31	5.22±.50
S. Kechil	8.39±.05	8.11±.18	8.39±.19	4.89±.48
Peta	5.67±.56	6.06±.50	7.72±.31	6.44±.41
B. E.-3	7.94±.31	7.78±.26	7.26±.33	6.61±.41

* Standard error.

followed by B.E.–3. The difference between the two varieties, however, was not significant. For the other varieties, significant flavor differences were noted at different cooking times.

Doneness.—The optimum cooking time for these varieties of rice ranged from 23 to 25 minutes. Raminad and S. Ketchil obtained higher scores, though insignificant than the other varieties. Raminad, however, was rated better done than Peta which obtained the lowest mean score.

SUMMARY AND CONCLUSIONS

Standardized procedures for cooking 400 grams rice were used and methods for measuring physical qualities and evaluating eating qualities are given. Results are reported for water absorption in washings, water uptake ratio, volume of cooked rice and palatability of cooked rice based on color, cohesiveness, off-flavor, and doneness.

Experiments showed that water absorption in washing had no significant effect on the cooking quality of rice. Water uptake

ratio is related to variety and the amount of cooking water used which is smallest with the least amount of cooking water. As the length of cooking time is increased, water uptake ratio decreased in all levels of cooking water due to evaporation of moisture.

Water uptake ratio decreases as rice ages as shown in Table 2 and Figures 1, 2, and 3 due to loss of moisture during storage. Long grain varieties showed slightly higher water uptake ratio at the first trial, while short grain had higher water uptake ratio at the third trial. Results indicate that as rice ages more water is needed particularly for the long grain variety.

The volume of cooked rice decreased as the length of cooking time increased. Volume is directly proportional to the amount of cooking water used. It was observed that there was an increase in volume as rice ages in S. Kechil while Raminad and Tjahaja showed a decrease in volume after storage.

Score cards for palatability test were used to determine the eating qualities of cooked rice. Mean scores showed that Raminad has better eating qualities based on color, cohesiveness, off-flavor, and doneness. B. E.–3 and Tjahaja ranked next in the order of preference.

The optimum cooking time for these varieties of rice ranged from 23 to 25 minutes.

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THE SELECTION OF DATA FOR BASAL METABOLIC RATE STANDARDS

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The basal metabolism of an individual is usually measured in the morning under conditions of complete physical and mental rest while the subject is awake, between 12 to 18 hours after a meal, and at a comfortable environmental temperature. These conditions are intended to standardize the procedure so that the rate of metabolism obtained is relatively constant and low. They merely seek to exclude the effects of muscular activity, mental and physical stress, specific dynamic effect of food, and extremes of environmental temperature. Consequently, the result obtained is not necessarily "basal," or the lowest for the individual, since lower values may be obtained under other conditions, such as profound sleep and chronic undernutrition. For this reason, Krogh(7) proposed the term "Standard Metabolism" to erase the erroneous impression that basal metabolism is the least resting metabolism, i.e., the lowest that could be obtained from an individual. This view was supported by other authors.(3, 4, 6, 12) Although the term "Standard Metabolism" has not gained universal usage, the meaning of basal metabolism has been accepted as such.

There is, however, considerable disagreement in the literature as to the manner of selecting data for constructing basal metabolic rate (BMR) standards. The variability of BMR values obtained from the same individual under supposedly identical standard conditions is well known to the researcher. As to which of the different values is to be regarded as the metabolic rate of the individual and used in the construction of standards has not been settled up to the present.

Boothby, et al.(3) pointed out that variability, being a universal biologic phenomenon, is inherent in metabolism values, and that "it must be taken into account by proper statistical methods and not excluded by predetermined selection of results." They maintained that there is not a single lowest value for basal metabolism for an individual since it is "a variable quantity that can be standardized but not absolutely fixed." They, there-

fore, constructed standards, now known as the Mayo Foundation Standards, from the first determination on all their subjects. Since this type of investigation calls for a large number of subjects, they claimed that it would be impractical to get the mean or the lowest of several determinations from the same subject because the values obtained by either method will depend upon the number of determinations: the more determinations, the lower will be the value obtained. Thus, the number of determinations should be strictly identical for each individual. This would necessarily limit the number of subjects in most investigations.

Robertson and Reid,(10) in constructing the British standards of basal metabolism, criticized the views of Boothby, et al. They claimed that the factor of nervous tension at the initial test is not uncommon, and that the initial reading is commonly higher than subsequent or true basal levels not only because of psychological factors but also because of training. Thus, they made duplicate determinations for several days until no further fall in the heat output was observed. The lowest reading obtained in the series was taken as the BMR value.

Webster, et al.,(14) in constructing standards of basal metabolism for adolescent Americans, performed two 6-minute observations and the average taken. If these two observations were not in agreement, the test was repeated until check observations were obtained. The report, however, failed to state within what degree of agreement an observation was considered acceptable.

Lamb and Michie,(8) in their study of the basal metabolism of children, made duplicate determinations on at least two mornings. This was continued until the lowest figure obtained was within 5 per cent of the results obtained on a different morning. The authors claimed that since basal metabolism cannot be assumed to be the single lowest figure but must consider any value within 5 per cent of the lowest, all figures checking within 5 per cent of the lowest were averaged with the lowest, and this was taken as the true basal metabolism of the individual.

In the Philippines, studies on the basal metabolism of Filipinos were conducted by Sison and Ignacio(12) and Ocampo, et al.(9) Sison and Ignacio performed duplicate tests for two successive days but did not state which figure was used for analysis. Ocampo, et al. performed duplicate tests on two

or three different days, and the lower value for any one day was taken as the basal metabolism value for that day. The average of these lower values for the different days was taken as the basal metabolism value of the individual.

The need for the establishment of BMR standards for Filipinos has long been recognized. For want of such standards, the Mayo Foundation Standards(3) have been used in clinical computations and in almost all other computations requiring basal metabolism values with adjustments made according to personal observations or past studies, such as those of Sison and Ignacio(12) and Ocampo, et al.(9)

For the purpose of establishing such standards, a group, composed of representatives of the National Science Development Board, Far Eastern University Dietary Department, University of the Philippines Home Economics Department, and the Food and Nutrition Research Center, was organized to make plans for a cooperative BMR study on Filipinos. The group agreed on standard methods of determining BMR by direct calorimetry, obtaining duplicate determinations on three successive days, and choosing, tentatively, the lowest of all the values obtained as the "true" value for the subject.

Because of disagreement on the method of selection of data for BMR standards in the literature cited above, a pilot study on 30 female subjects was done by the FNRC to test the methods agreed upon by the study group. Various methods of selection of data were tried and the results compared with one another to determine those which could serve as guide in the study and establishment of BMR standards for Filipinos.

METHODS

Thirty females, aged from 19 to 26 years, and all within 10 per cent of their standard weight, were taken as subjects of this study. They were all adjudged normal by physical examination and were negative for any history or sign of thyroid or other diseases that might alter basal metabolism. On the day before the tests, the subjects were given a preliminary test with use of the Benedict-Roth metabolism apparatus in order to acquaint them with the instrument and with the conditions required by the experiment. They were then instructed to take a light evening meal, to fast, and to avoid strenous physical activity.

The following morning they reported directly to the clinic with a minimum of activity. Benedict and Crofts(1) have shown that this amount of activity (which includes dressing, walking and climbing stairs) does not materially increase the basal heat production as long as the subject rests in bed for 30 minutes before the test.

Weight and height were taken with ordinary light clothing and without shoes. The weight was later adjusted to nude weight for the purpose of surface area determinations by subtracting 0.6 kg from this figure. Six-tenths kilogram was found by preliminary trials as the average weight of clothes worn by women in this age group. Relatively large errors in this figure do not significantly affect surface area. The subjects then rested in bed from 30 minutes to one hour, and their temperature, pulse rate, and blood pressure were recorded immediately before the test.

Oxygen consumption was determined by means of the Benedict-Roth metabolism apparatus, which was tested occasionally for leaks and for accuracy in the rate of rotation of the drum. Two successive 8-minute tests were run at an interval of about 5 minutes. Results were expressed in calories per square meter of body surface per hour.

The tests were repeated for three consecutive mornings; so that, in all six determinations consisting of three duplicates were made on each subject. Results were discarded when the subject showed obvious signs of tension, or when leaks occurred during the test.

The data from all accepted readings for the 30 subjects were then segregated by four main methods of selection; and the results obtained by these four methods were compared not only with one another but with those obtained by other methods. The four methods, chosen because of their distinct characteristics and because they were popularly used by other authors, were:

(1) Mean of all determinations (the BMR of the subject was taken as the mean of all determinations taken of the subject),
(2) First determination (only the first observation was considered),
(3) Lowest of all determinations (only the lowest value was chosen), and
(4) Mean of the lower of three duplicates (the lower values in each of the three days were averaged and taken as the BMR of the subject).

The other methods, which are variants of the four above, were:

(1) Mean of the first duplicate (the observations of the first day were averaged),
(2) Mean of the first four determinations (all duplicate determinations on the first two days were averaged),
(3) Mean of the first of two duplicates (only the first determinations in each of the first two days were averaged),
(4) Mean of the first of three duplicates (the first determinations in each of the three days were averaged),
(5) Lower of the first two determinations (only the lower value on the first day was considered),
(6) Lowest of the first four determinations (only the lowest of all values on first two days was considered), and
(7) Mean of the lower of the first two duplicates (the lower values in each of the first two days were averaged).

The data were also analyzed more minutely by comparing results within the day and from day to day. Further selection of cases was done by getting only values checking within 5 per cent of their duplicates, or values checking within 5 per cent of the lowest for each individual series.

Statistical comparisons were then made mainly by determining the t-distribution of paired differences between results obtained from the various methods.

RESULTS

Table 1 gives the mean, standard deviation, and coefficient of variation of the basal metabolism of the 30 subjects obtained by the different methods of selecting data. No attempt was made to compare the means obtained from the different methods with one another nor with established standards since the age of the group studied was not as homogeneous as was necessary.

TABLE 1.—*Basal metabolism of thirty female subjects according to the different methods of selecting data.*

Methods of selection of data	Mean	Standard deviation	Coefficient of variation
	Cal/m²/hr	*Cal/m²/hr*	*Per cent*
1. Mean of all determinations	34.96	2.34	6.69
2. Mean of first duplicate	35.10	3.82	10.88
3. Mean of first 4 determinations	35.02	2.59	7.36
4. Mean of first of 2 duplicates	35.29	3.75	10.63
5. Mean of first of 3 duplicates	35.25	2.70	7.65
6. First determination	35.17	4.46	12.68
7. Lowest of all determinations	31.65	2.53	7.99
8. Lower of first 2 determinations	33.95	4.06	11.69
9. Lowest of first 4 determinations	32.12	2.88	8.69
10. Mean of lower of 3 duplicates	33.82	2.08	6.15
11. Mean of lower of first 2 duplicates	33.78	3.02	8.94

It will be noted, however, that the standard deviations, except in a few specified instances, are comparable with those obtained by other authors.(2, 3, 10, 15) Consequently, the coefficients of variation obtained with the use of the different methods of selection are all within the accepted limit of 10 per cent, with four exceptions; namely,(1) mean of the first duplicate,(2) mean of the first of two duplicates,(3) first determination, and(4) lower of first two determinations. In other words, when only one or two readings out of six are considered, the standard deviations of the results are rather high.

Table 2 shows the comparison of the results. The four principal methods of data selection were compared with one another and with other methods. The difference between the mean of all determinations on each subject and either the lowest of all determinations or the mean of the lower of the three duplicates was found highly significant. There was, however, no statistically significant difference between the mean of all determinations and either the first determination, the mean of the first duplicate, the mean of the first four determinations, the mean of the first of two duplicates, or the mean of the first of three duplicates.

Similarly, the first determination was neither significantly different from the mean of the first duplicate, the mean of the first four determinations, the mean of the first of two duplicates, and the mean of the first of three duplicates. The first determination was, however, significantly different from the lowest of all determinations and from the mean of the lower of three duplicates.

When the lowest of all determinations was selected, the picture was very different. The results obtained by this method were significantly different from the results obtained by the other methods with which they were compared. Thus, the difference between the lowest of all determinations and either the mean of all determinations, the first determination, the lower of the first two or lowest of first four determinations, the mean of the lower of three duplicates or the mean of the lower of first two duplicates was significant when tested statistically.

On the other hand, the mean of the lower of three duplicates was not significantly different from either the lower of the first duplicate or the mean of the lower of the first two duplicates. It was, however, significantly different from the

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TABLE 2.—*t-Distribution of paired differences between results obtained by different methods of selection of BMR data.*

Methods of selection of data	Mean of all determinations	First determination	Lowest of all determinations	Mean of lower of 3 duplicates
	t(p)	t(p)	t(p)	t(p)
1. Mean of all determinations		0.3170 (>.70)	12.0119 (< .001)**	5.8316 (<.001)**
2. Mean of first duplicate	0.2864 (>.70	0.7045 (>.40		
3. Mean of first 4 determinations	0.2980 (>.70	0.4221 (>.60		
4. Mean of first 2 duplicates	0.9038 (>.30	0.1283 (>.80		
5. Mean of first 3 duplicates	0.9618 (>.30	0.2105 (>.80		
6. First determination	0.3150 (>.70		5.8494 (<.001)**	2.1797 (<.05)*
7. Lowest of all determinations	12.0119 (<.001)**	5.8491 (<.001)**		9.9862 (<.001)**
8. Lower of first 2 determinations			6.2504 (<.001)**	0.3099 (>.70)*
9. Lowest of first 4 determinations			2.2866 (<.05)**	5.4540 (<.001)**
10. Mean of lower of 3 duplicates	5.8316 (<.001)**	2.1797 (<.05)*	9.9862 (<.001)*	
11. Mean of lower of first 2 duplicates			2.1040 (<.05)*	0.3063 (>.70)

* Significant
** Highly significant

lowest of first four determinations, and, as has already been mentioned, the mean of all determinations, the first determination and the lowest of all determinations.

When the mean of the duplicates was considered, 64 per cent of the cases had a lowering of their BMR values from the first to the second day, but 50 per cent showed a return towards the original value from the second to the third day. Although there were occasional marked inconsistencies in the results during the day and from day to day in the whole series, the differences were not marked in most of the cases. Thus, in general, there were as many cases that showed a rise in their BMR values from the first to the second determinations on the same day as there were that showed a fall. In only a few instances were the results on the same day the same.

However, when data were selected in such a manner that only those values that agreed within 5 per cent of their duplicates were considered, only 40 per cent of the cases were included, eliminating the rest when only one duplicate was considered sufficient for analysis. If it were necessary to make the duplicate agree within 5 per cent on at least two days, two-thirds of the data would be eliminated. In no case did the duplicates agree within 5 per cent in all three days. When only figures checking within 5 per cent of the lowest for each individual were used in the analysis, only 53 per cent of the cases were included.

DISCUSSION

It is recognized that the statistical methods used in the analysis of the data merely serve to show whether one method of selection gives results which are, with greater or lesser probability, the same or different from one another. In other words these statistical methods cannot, by themselves, make one choose which method of selection is the correct one among those tested. However, either of two methods whose results are found not to be significantly different may be used with a high probability of being correct as long as the same conditions are met.

The number of subjects used in this study was chosen because more or less the same number for each age group will be used for the construction of standards. This number was deemed sufficient since the standards will be based, as it should be, not on straight averages of each age group, but on

statistical methods which consider the behavior of all age groups. This method was used by Boothby, et al.(3) and Robertson and Reid(10) in their construction of BMR standards, where, incidentally, about 30 subjects or less were included in each age group.

Of the four main methods of selection of data used in this study, those that involved selective sampling schemes (lowest of all determinations and mean of lower of three duplicates) were significantly different from those that did not (mean of all determinations and first determination). Thus, the mean of all determinations was not significantly different from the first dtermination, the mean of the first duplicate, the mean of the first four determinations, the mean of the first of two duplicates, or the mean of the first of three duplicates. In other words, one may choose from any of these six methods without materially affecting the results if it were decided that no selective sampling scheme should be employed.

However, it was shown that when only one or two observations out of six were considered, (first determination, the mean of first duplicate, the mean of first of two duplicates), the standard deviations of the results were rather high. It would, therefore, not be safe to employ these methods for the construction of standards, especially when the number of subjects for each age group is less than the number studied here. It is pointless to consider the mean of the first of three duplicates since it would not offer any advantage over getting the mean of two-day observations. It appears that getting the mean of all determinations made for two consecutive days is sufficiently complete to give a representative value for an individual.

Two arguments against such a scheme are: (*1*) the element of training is absent in the first day, causing a generally lower observation on subsequent days; and (*2*) certain factors, such as slight nervous tension, mental irritations, etc., being unappreciated by the investigator, may cause a slight rise in metabolism in spite of the apparent fulfillment of all conditions for BMR determinations.

The element of training has been considered by several observers(5, 10, 11, 13, 14) as an important factor in BMR determinations. It seems, however, that this factor was obviated in the present study because of the preliminary test conducted on each subject a day before the series was begun. When the means of the day's observations were considered, the first day's results did not differ significantly from those of the second day.

The possibility that such factors as nervous tension, mental irritation, and apprehension may cause a rise in metabolism cannot be ignored. The investigator may not be so sufficiently aware of their presence that erroneous results are accepted and errors introduced when simple averages are used in the selection of data. Indeed, this had happened not infrequently in the course of the present study: some values were too obviously different to be considered part of biological variability. The rise in the BMR observed in the third day was most probably due to irritation from repeated tests, similar to what Berkson and Boothby(2) had observed. Theoretically it would be possible to recognize such errors at the time of the test, but it is really difficult to do so. It, therefore, would be more accurate in this case to select either the lowest of all observations or the mean of the lower of several duplicates.

The objections of Boothby, et al.(3) against the use of the lowest of all determinations have already been mentioned. The use of this value in the construction of physiological standards disregards the factor of variability which is inherent in such standards. The results obtained will therefore reflect the floor of the measurement, and most individuals compared with this standard, as in clinical use, will necessarily fall above this value. Since the level of the lowest value will depend upon the number of observations, all determinations, either in the construction of standards or for purposes of comparing with the standards, should have an identical number of tests. Otherwise, as this study has shown, the results may be different.

On the other hand, the mean of the lower of several duplicates takes into account the factor of variability. The method follows the concept that there is no single fixed value of basal metabolism for an individual. Also, by getting only the lower values, most of the observations that have been vitiated by unrecognized extraneous factors are eliminated, so that more consistent results are obtained. This was confirmed by the observation that the mean of the lower of three duplicates had the lowest coefficient of variation among the various methods used in the present study (Table 1).

Since it was also observed that the mean of the lower of three duplicates was not significantly different from the mean of the lower in each of two days' observations, the latter may be used without materially affecting the results. It is felt

that whatever small difference there is between the results of these two methods is offset by the advantages offered by the shorter duration of tests when only two duplicates are obtained.

SUMMARY

A pilot study of the basal metabolism of 30 female subjects was done in order to compare various methods of selecting data for the establishment of BMR standards for Filipinos. Six measurements, consisting of duplicate measurements of oxygen consumption for three successive days, were made on each subject under basal conditions using the Benedict-Roth metabolism apparatus. It was shown that when only one or two readings out of six were considered, the standard deviations of the results were rather high. It would not be safe therefore, to use these methods for the construction of standards when the same or less number of subjects is employed.

The chief argument cited against the use of the mean of two or three days' observations is the possibility that extraneous factors, unappreciated by the investigator may cause a rise in metabolism, thereby introducing errors when simple averages are used. Objections against using the lowest of all determinations were also reviewed.

Getting the mean of the lower of several duplicates (at least two duplicates) is probably the best under the conditions of the present study because this method not only accounts for the factor of variability but also eliminates readings that are vitiated by unrecognized extraneous factors. A preliminary test on each subject is, however, necessary to prevent high initial readings due to apprehension and absence of training.

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POSSIBLE USE OF ELECTRIC CURRENT FOR INCREASING VOLUMETRIC RATE OF FLOW OF OIL AND WATER DURING PRIMARY OR SECONDARY RECOVERY

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TWO PLATES AND TWO TEXT FIGURES

INTRODUCTION

Historical background.—The earliest recorded experiments with electro-osmosis were conducted by Reuss, who discovered the phenomenon in 1808 [Reuss (1809) 327]. Reuss also discovered the electrophoresis phenomenon in the course of his experiments.

The first quantitative experiments dealing with electro-osmosis were conducted by Wiedemann in 1852 [Wiedemann (1852) 321)]. Wiedemann discovered that the rate of water transport by electro-osmosis through clay diaphragms was a function of current and is independent of the geometry of the porous material. Wiedemann achieved considerable accuracy in his experiments.

In 1859, Quincke proved that the magnitude of the potential generated by forcing water through porous material was independent of the dimensions of the material, but did depend on the nature of the material and the applied pressure across the material [Quincke (1859) 1]. He proposed a rigid "double charge layer" theory based on his findings that most surfaces acquire a negative charge when wet with water. According to Quincke a layer of positive charges was attracted from the liquid to counterbalance the negative surface charges.

Theory.—The difference in potential ζ (zeta potential) between the layers of charges (−) affixed to the particles and the layer of balancing charges (+) in the liquid bathing the solid particles is equal to Q/C where Q is the charge and C is the capacity per unit area of the capacitor, which is in-

versely proportional to the distance between the solid particles and the charge layer in the liquid. Zeta potential ζ depends on the type of substance in contact and on the diameter of capillaries.

If the applied electrical field having intensity ε is parallel to the axis of capillaries, then force $F=Q\varepsilon$ would move the charged layer of fluid (and the adjacent fluid) with a velocity u_e. This velocity depends mainly on the size of capillaries, viscosity of the fluid, dielectric constant, and zeta potential. Thus, $u_e=f\triangle\varepsilon$ where f is the proportionality factor reflecting the aforementioned factors, and $\triangle\varepsilon$ is the gradient of the electric potential. If velocity due to the hydrostatic head is u_h, then the total velocity is the sum of the u_e and u_h. This force might aid the hydrodynamic forces in forcing the water into the formation, and the efficiency of this method will probably depend on the concentrations of clays in the sandstones.

EXPERIMENTAL PROCEDURE AND APPARATUS

Method of coring—The unconsolidated mixture is placed in a 1-inch o.d. polyethylene tubing 15 cm in length. The tubing is first stoppered on one end; and then the filled tubing is tapped several times until a sufficiently tight packing of the mixture is obtained. On each end, an alundum coarse disk No. 282–75 (Braun) is placed between the rubber stoppers and the mixture. The disks are included to serve as filters, whereas the purpose of the rubber stoppers are to leave a vacant space after the epoxy resin has dried.

The stiff sausage-type tubing (Plate 1, fig. 1A) is then placed in a vertical collar sleeve (Plate 1, fig. 1B) which has been made from gauge 30 copper sheet, and the epoxy resin (Hysol) is poured. It is preferable, however, to pre-grease the inside surface of the sleeve so as to facilitate its removal after the epoxy has dried. The sleeve is then placed in the pressure bomb (Plate 2) and subjected to the desired pressure. The core is left in the pressure chamber for 24 hours. This method has been found to be best because in the pressure chamber all forces acting on the sleeve body are equal insuring a good compaction, and eliminating any form of skin or chanelling effects at the contact of the tubing and mixture surfaces. Eight threaded holes are drilled on each core to be used for external attachments (Fig. 1).

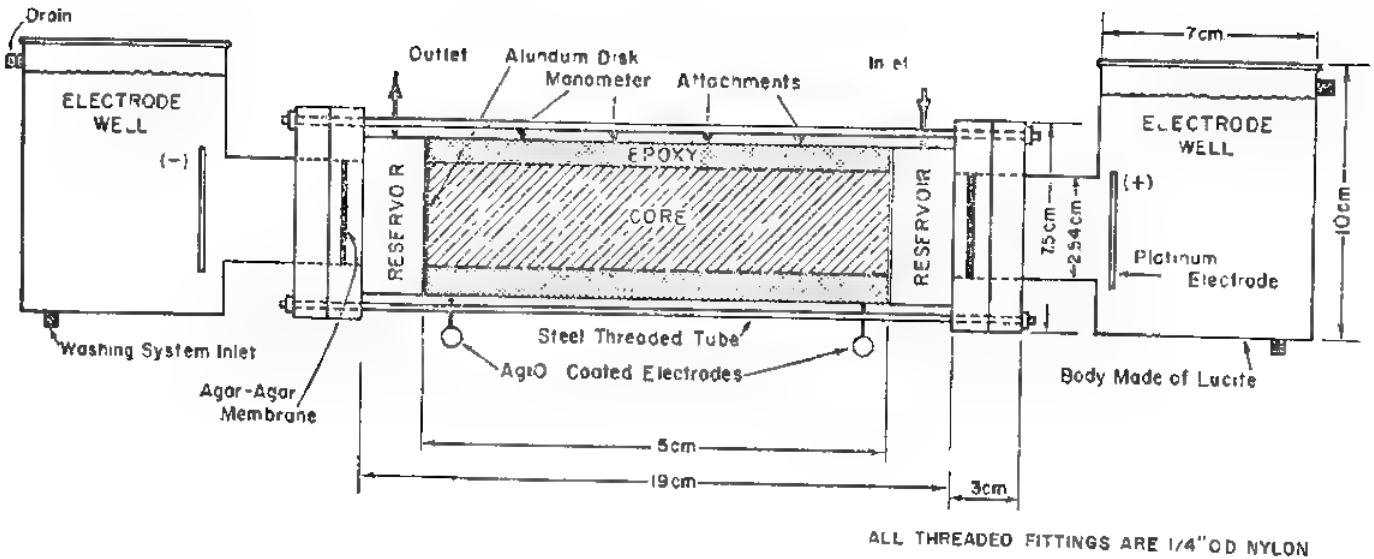

FIG. 1. Schematic diagram of apparatus.

Description of equipment.—Four manometers were used to observe the pressure gradient at equal intervals and also to indicate any swelling of the clays (Plate 1 and Fig. 2). Mercury was used in the manometers; and in order to eliminate any reaction between the mercury and the inlet solution (1 per cent by wt. NaCl solution plus 150 ppm formaldehyde), a kerosene phase was used between the mercury and the inlet solution.

The imposed current was supplied by a D. C. voltage rectifier. Two platinum electrodes were used which were immersed in 1 per cent by wt. NaCl solution (Fig. 1). Two coarse alundum disks (No. 282–75 Braun) were treated with a light solution of agar-agar and were used as the membranes (Fig. 1). The inlet solution from the tank was filtered through a fine alundum disk (No. 282–76 Braun) preceded by two strips of filter paper to assure a debris-free inlet solution, in the case that some foreign matter might have been introduced through the compressed air system.

To eliminate any polarization effects, a washing system was devised so as to continuously introduced a fresh-saline solution into the platinum-electrode well. Two Ag_20 (Ag–AgCl) coated electrodes were used to measure the potential drop across the core.

EXPERIMENTAL RESULTS

The experimental conditions and results are presented in Tables 1 and 2.

TABLE 1.—*Experimental conditions.*

Core No.	Composition	Mesh (Tyler)	P coring (psig)	P inlet (formation water) (psig)
	Per cent			
I	95 Silica 5 montmorillonite clay	325 400	1000	15
II	95 Silica 5 montmorillonite clay	325 400	800	15
III	95 $CaCO_3$ 5 montmorillonite clay	325 400	1000	5

TABLE 2.—*Electrical potential imposed versus (volumetric rate of flow)/(initial flow) ratio.*

Core No.	Potential, volts	Flow, (cc/min) x 10³	q/q₁
I	0.0	3.13	1.000
	25.0	3.70	1.180
	25 0	3 71	1.184
	27 5	3 84	1 225
	50 0	4 43	1 420
	50.0	4.76	I 520
	00 0	4.94	1.577
	75.0	5.10	1.630
	75 0	5.13	1.639
	75 0	5 14	1 640
	100 0	5 14	1 640
	100 0	5.15	1 645
	100 0	5.16	1.650
II	0 0	38.80	1.000
	50 0	40 50	1.045
	63 5	41 50	1 070
	65 0	40 60	1 145
	6 . .	48.50	1 250
	66 0	50.60	1.303
	70.0	52 40	1 350
	70 0	53.20	1.370
	75 0	53 10	1 375
	100 0	53.70	1 388
	101 0	54 40	1 400
	104.0	5 . 90	1.449
	105 0	57.09	1.470
	106.0	57.20	1.472
III	0 0	89 00	1 000
	7.0	102 00	1 145
	7 .	98 30	1 10 .
	18.0	10 . 00	1.180
	18.2	102 50	1.152
	22 5	108.00	1.215
	24 0	106.80	1.200
	24 5	110.00	1.235
	46.6	134 20	1.510
	46 0	133.00	1.500
	18 0	127.80	1.435
	64 0	164.50	1.850
	65 0	172.50	1.940
	81 0	201.00	2.260
	50 0	192.50	2.160
	92 0	208.00	2.340

Steady state hydrodynamic flow: Core I: 3.13×10^{-3} cc/min
Core II: 38.80×10^{-3} cc/min
Core III: 89.00×10^{-3} cc/min

As shown in Tables 1 and 2 and Fig. 2, the volumetric rate of flow increases with increasing electrical potential imposed.

EXPERIMENTAL ACCURACY

There are several sources of error in the experimental procedure used, and most of these are fairly easy to estimate. In the measurements of the cell constant, a probable maximum error is about 4 per cent. In the measurements of time and volume for permeability determination, the maximum error in each case is estimated at 1 per cent. Error in voltage measurements does not exceed 3 per cent.

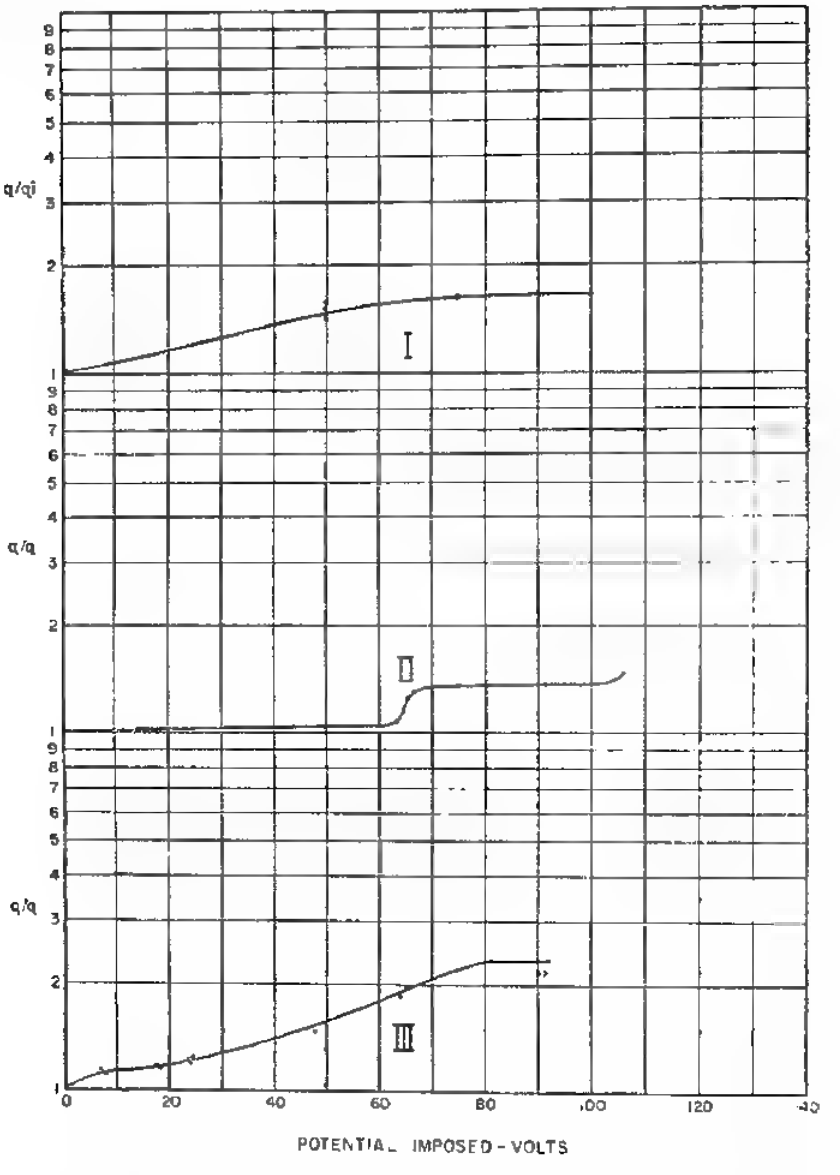

FIG. 2. Relationship between (volumetric rate of flow)/(initial flow) ratio and imposed voltage. Also see Tables 1 and 2.

I. Silica with 5 per cent by weight of montmorillonite clay.
II. Silica core with 5 per cent weight of montmorillonite clay. The initial voltage applied in this case was 50 volts.
III. $CaCO_3$ core with 5 per cent by weight of montmorillonite clay.

CONCLUSION

There are possibilities for applying this method in providing a process for enhancing the injectibility of water in water flood systems and also to provide a process combining electrical effects and chemical effects so as to increase the injectibility of the formation in water injection processes, or simply increase the rate of production in tight formations. This may be done by the introduction of an electrode (such as graphite or carbon) into each well at the depth of the formation being flooded, and connect an electrical generator with the two electrodes, so that the electrode in the injection well is positive and the electrode in the producing well is negative. The applied voltage may be programmed so it will be, for example, applied each hour at about 100 volts, for quarter hour, the 45 minutes of each hour remaining without any voltage applied at all. Numerous variations are possible as to their usefulness in a particular case, and will be found to vary from one installation to another. Water is pumped into the injection well in the usual fashion. This investigation gives evident proof that when the electrical voltage is applied to the electrodes, the injection rate immediately increases; all other conditions of pump pressures and the like being constant. Also, the use of chemical additives in low concentrations (0.01 to 0.1 g/100 cc) will increase the effectiveness of this process even though they may entail some reduction in resistivity. As shown by Tchillingarian (1952), negative ions adsorb on the clay particles and thus impart a greater negative charge to clay.

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ILLUSTRATIONS

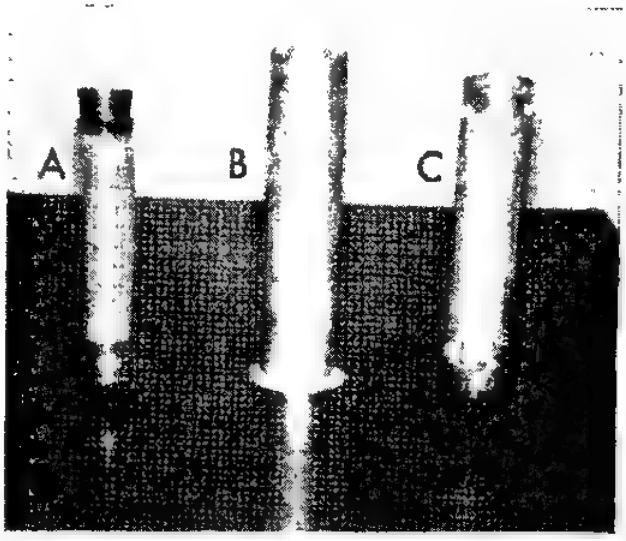
PLATE 1

FIG. 1. Photograph of coring steps: A, Sausage-type polyethylene tubing filled with unconsolidated mixture tightly packed and stoppered; B, Copper sleeve; C, Complete core to be used for experiment.

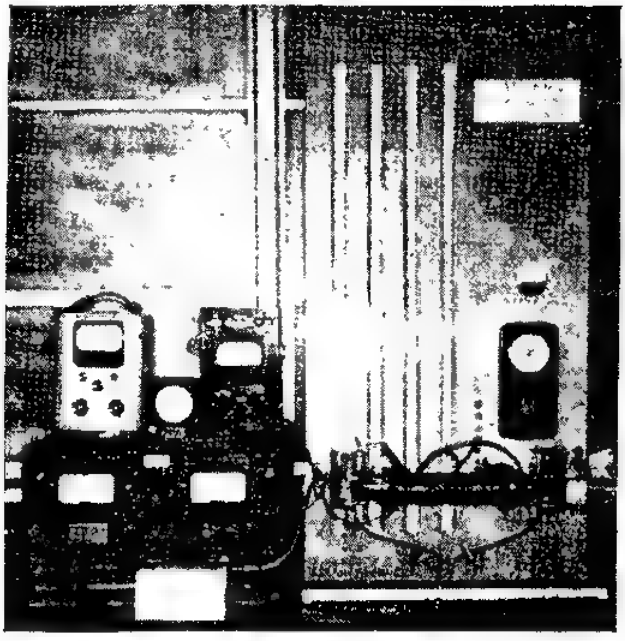
2. Photograph of complete unit.

PLATE 2

Photograph of pressure bomb.



1



2

PLATE 1.

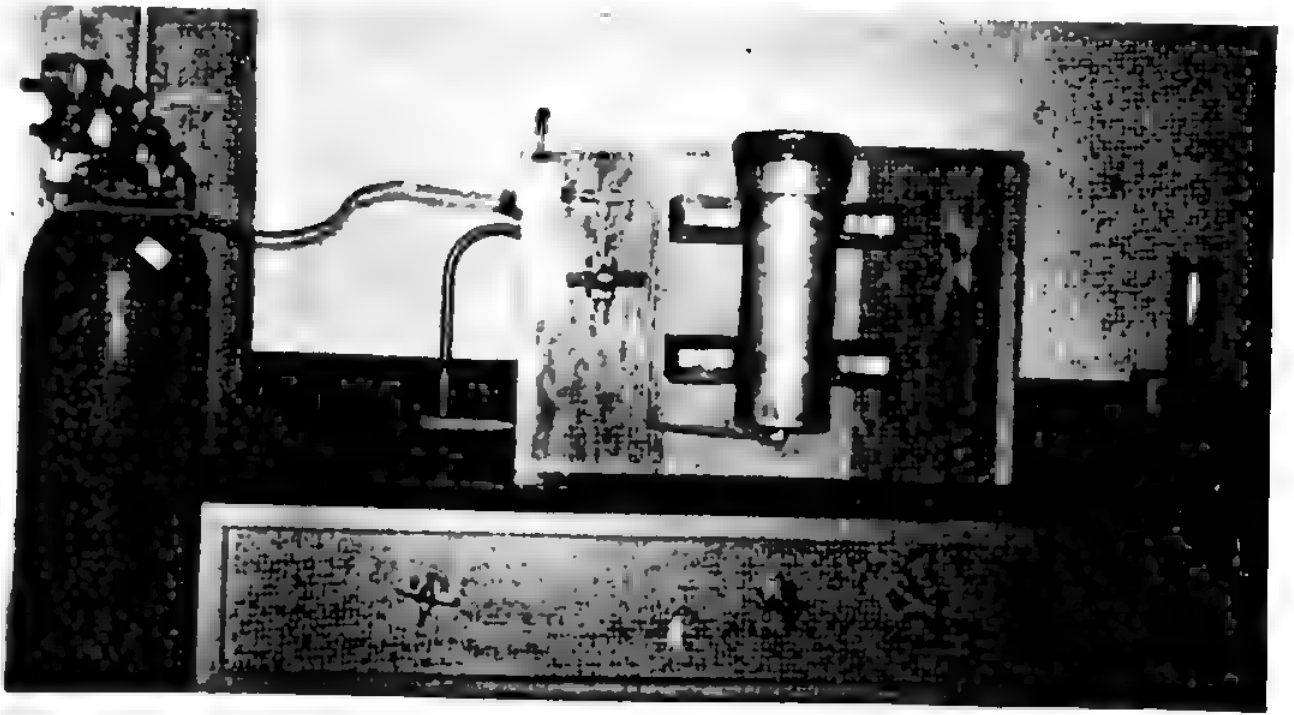


PLATE 2.

PHYSICS CALCULATIONS FOR THE PHILIPPINE OPEN-POOL REACTOR PRR-I. C. REACTOR CONTROL

By JOSE O. JULIANO and CESAR L. PINEDA
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THREE TEXT FIGURES

A thorough knowledge of the reactor control of PRR–I is a very important aspect in the safety evaluation of the Philippine Atomic Research Center. It is therefore, necessary to study and calculate the reactivity worths of the control blades and rod of PRR–I with available theoretical methods prior to the loading of the reactor. The control system of PRR–I is composed of four control blades and one servo-controlled regulating element.(1) The former are intended for coarse control of the reactor like shut-down and compensation for major reactivity changes, while the latter is intended for small reactivity changes. The position of the control blades and regulating element in the core is given in Fig. 1. Each control blade, 54 ⅛″ × 10 ⅝″ × ⅜″, is made of Boral sandwiched between two aluminum sheets, while the servo-controlled element is a 2⅛″ square tube made out of ¼″ thick Boral sheet cladded with 40-mil aluminum.

In this report two methods, both based on neutron diffusion theory, were used to calculate the blade control-worths of the PRR–I.

METHOD A

The first method is patterned after Haling, et al.(2) with some modifications and corrections. The following assumptions are made:

1. Neutrons in the core are divided into two energy groups; namely, thermal with 0 to 0.625 ev energies, and fast neutron group with energies above 0.625 ev.
2. The control blades are black to thermal neutrons while transparent to fast neutrons.
3. The control blades are extremely thin.
4. The PRR–I core is a bare homogeneous parallelepiped core with the effect of the reflectors incorporated in the reflector savings.
5. The current boundary condition satisfied at the blade surface is:

$$\frac{d\phi_2}{dx} = \frac{1}{\delta}\,\phi_2$$

where δ is the slow-neutron extrapolation distance and ϕ_2 is the thermal flux.

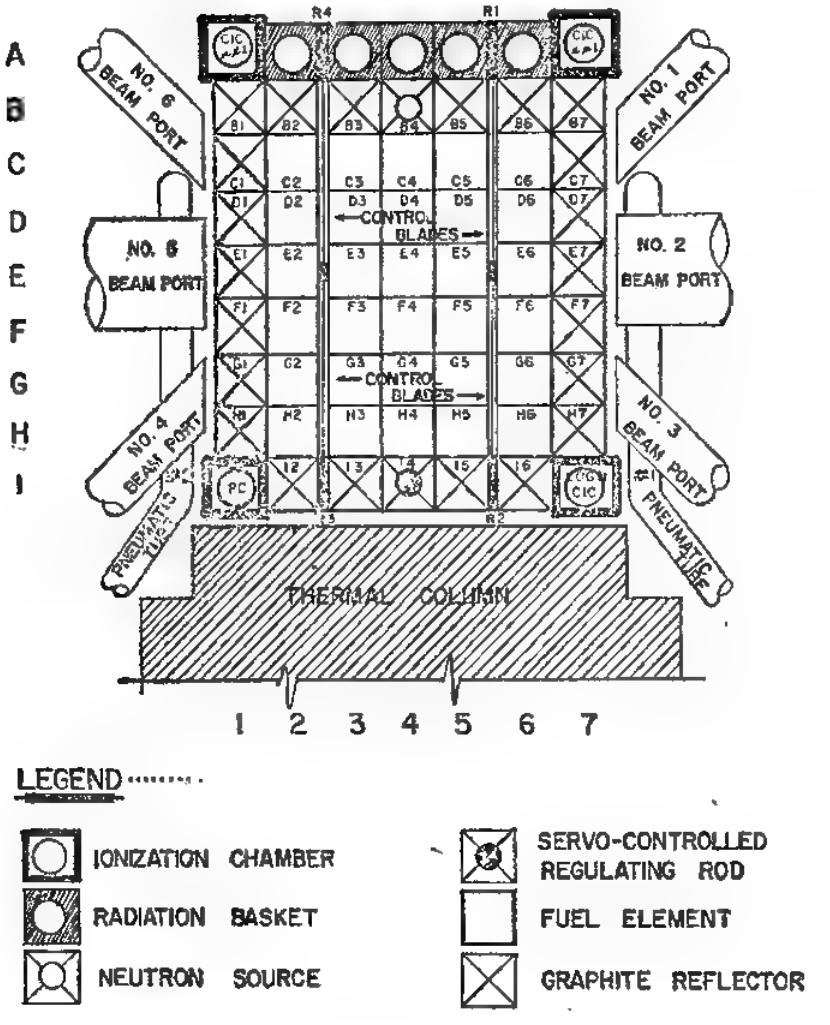

FIG. 1. PRR-I core and control system positions.

6. The core constants of Juliano(3) are assumed to represent the true characteristics of PRR-I.

In this method two cases were studied; namely, first when all the four control blades are inserted into the core and second, when two adjacent control blades are inserted and the other two adjacent blades fully withdrawn from the core.

The diffusion equations solved were

$$\Delta^2\phi_1 - \frac{1}{\tau}\phi_1 + \frac{k\Sigma_2}{D_1}\phi_2 = 0 \tag{1}$$

$$\nabla^2\phi_2 - \frac{1}{L^2}\phi_2 + \frac{\Sigma_1}{D_2}\phi_1 = 0 \tag{2}$$

where the solutions are assumed to satisfy

$$\nabla^2\phi + B^2\phi = 0 \tag{3}$$

The nontrivial solutions of (3) are

$$B_1^2 = \tfrac{1}{2}\left[-\left(\frac{1}{\tau}+\frac{1}{L^2}\right) + \sqrt{\left(\frac{1}{\tau}+\frac{1}{L^2}\right)^2 + \frac{4(k-1)}{\tau L^2}}\right] \tag{4}$$

$$B_2^2 = \tfrac{1}{2}\left[-\left(\frac{1}{\tau}+\frac{1}{L^2}\right) - \sqrt{\left(\frac{1}{\tau}+\frac{1}{L^2}\right)^2 + \frac{4(k-1)}{\tau L^2}}\right] \tag{5}$$

It is assumed that the flux ϕ (x, y, z) can be represented by

$$\phi(x, y, z) = \phi(x)\,\phi(y)\,\phi(z) \tag{6}$$

$$\text{where } \phi(y)\,\phi(z) = \cos \alpha y \cos \beta z \tag{7a}$$

and

$$\alpha = \frac{\pi}{W} \quad , \quad \beta = \frac{\pi}{h} \tag{7b}$$

The general solution for $\phi(x)$ can be written as

$$\phi(x) = A \sin \mu x + E \sinh \nu x + \cos \mu x + D \cosh \nu x \tag{8}$$

where $\alpha^2 + \beta^2 + \mu^2 = B_1^2$

$$\alpha^2 + \beta^2 - \nu^2 = B_2^2$$

For the fast and thermal fluxes the corresponding general expressions are

$$\phi_1 = (A_1 \sin \mu x + E_1 \sinh \nu x + C_1 \cos \mu x + D_1 \cosh \nu x) \cos \alpha y \cos \beta z \tag{9}$$

$$\phi_2 = (A_2 \sin \mu x + E_2 \sinh \nu x + C_2 \cos \mu x + D_2 \cosh \nu x). \cos \alpha y \cos \beta z \tag{10}$$

Substituting (9) in (1) and equating coefficients of similar terms, the following relationships are obtained

$$A_2 = \frac{A_1 D_1 (B_1^2 + \frac{1}{\tau})}{k\Sigma_2} \tag{11}$$

$$C_2 = \frac{C_1 D_1 (B_1^2 + \frac{1}{\tau})}{k\Sigma_2} \tag{12}$$

$$E_2 = \frac{B_1 D_1 (B_1^2 + \frac{1}{\tau})}{k\Sigma_2} \tag{13}$$

$$D_2 = \frac{D_1^2 (B_2^2 + \frac{1}{\tau})}{k\Sigma_2} \tag{14}$$

Writing ϕ_2 in terms of A_1, E_1, C_1, D_1, and ϕ_1 by using equations (11) to (14) and dropping the subscript "1,"

$$\phi_1 = (A \sin \mu x + E \sinh \nu x + C \cos \mu x + D \cosh \nu x) \cos \alpha y \cos \beta z \tag{15}$$

$$\phi_2 = \left\{ \frac{AD}{k\Sigma_2} (B_1^2 + \frac{1}{\tau}) \sin \mu x + \frac{ED}{k\Sigma_2} (B_1^2 + \frac{1}{\tau}) \sin h\nu x + \frac{CD}{k\Sigma_2} (B_2^2 + \frac{1}{\tau}) \cos \mu x + \frac{D^2}{k\Sigma_2} (B_2^2 + \frac{1}{\tau}) \cosh \nu x \right\} \cos \alpha y \cos \beta z \tag{16}$$

From Fig. 2 it is seen that the boundary condition to be followed are:

1. At $x_a = 0$, $\phi_1 = \phi_2 = 0$, and this requires $C_a = D_a = 0$

2. At $x_b = 0$, $\frac{d\phi_1}{dx} = \frac{d\phi_2}{dx} = 0$, and this requires $A_b = E_b = 0$

3. At $x_a = a$ and $x_b = b$, $\phi_{1a} = \phi_{1b}$, $\frac{d\phi_{1a}}{dx_a} = -\frac{d\phi_{1b}}{dx_b}$

$$\phi_{2a} = -\frac{\delta d\phi_{2a}}{dx_a} \text{ and } \phi_{2b} = -\frac{\delta d\phi_{2b}}{dx_b}$$

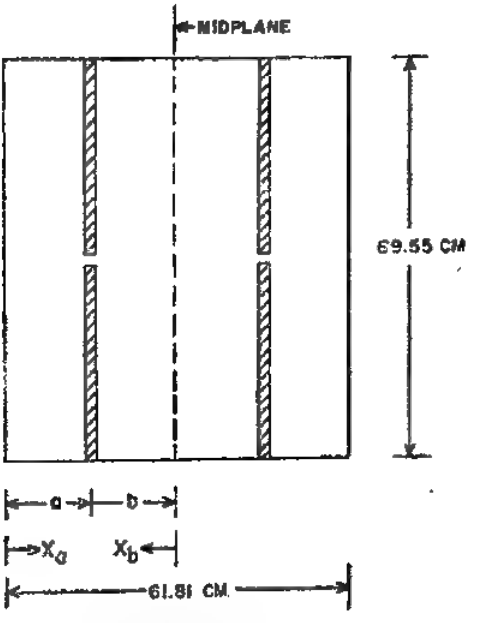

FIG. 2. Schematic diagram of PRR-I core with four blades inserted.

Upon using the above boundary conditions the final equation for the core becomes

$$\left\{\frac{1 - \frac{\upsilon}{\mu\Delta}\sin\mu a\left(\frac{1+\upsilon\delta}{\sin\mu a + \mu\delta\cos\mu a}\right)}{1 - \frac{\upsilon}{\mu\Delta}\cos\mu a\left(\frac{1+\upsilon\delta}{\sin\mu a + \mu\delta\cos\mu a}\right)}\right\} = \tag{17}$$

$$-\left\{\frac{1 + \frac{\upsilon}{\mu\Delta}\cos\mu b\left(\frac{1+\upsilon\delta}{\cos\mu b - \mu\delta\sin\mu b}\right)}{1 + \frac{\sin\mu b}{\Delta}\left(\frac{1+\mu\delta}{\cos\mu b - \mu\delta\sin\mu b}\right)}\right\}$$

where $\Delta = \frac{\upsilon}{\mu}\frac{(B_1^2 + \frac{1}{\tau})}{(B_2^2 + \frac{1}{\tau})}$; $\tanh \upsilon a$, $\coth \upsilon a$, $\tanh \upsilon b$, and $\coth \upsilon b$ have been assumed unit.

By assigning values to k, the values of the left hand side of (17), L, are calculated and plotted against k, likewise for the right hand side of (17), R. The intersection of curves of L and R gives the k value required.

The results for the condition when the four control blades are inserted in the core are tabulated in Table 1, a control worth of 19.46 per cent being obtained.

TABLE 1.—*Method A: Four control blades inserted.*

Constants used;

$a = 19.304$ cm	$b = 11.601$ cm
$\tau = 50.7793$ cm^2	$1/\tau = 0.01969$ cm^{-2}
$L^2 = 7.0143$ cm^2	$\delta = 0.784$ cm
$\alpha^2 + \beta^2 = 0.00367$ cm^{-2}	$B^2 = 0.00625$ cm
$k_{rodded} = 1.6427$	$k_{unrodded} = 1.3751$

worth of four control blades = 19.46 per cent

k	D_1^2	D_2^2	μ^2	ν^2	L	R
1.640	0.01040	−0.17266	0.00674	0.17633	7.9070	8.1004
1.645	0.01048	−0.17274	0.00682	0.17611	7.9955	7.8385
1.650	0.01056	−0.17282	0.00689	0.17649	8.0856	7.5943

When two adjacent control blades are fully inserted the new boundary conditions inferred from Fig. 3 are:

1. At $x_a = 0$, $\phi_{1a} = \phi_{2a} = 0$
   $x_b = 0$, $\phi_{1b} = \phi_{2b} = 0$ } satisfied by $C = D = 0$
2. At $x_a = a$, $x_b = b$, the conditions are

$$\phi_{1a} = \phi_{1b} \qquad \phi_{2a} = -\frac{\delta d\phi_{2a}}{dx_a}$$

$$\frac{d\phi_{1a}}{dx_a} = -\frac{d\phi_{1b}}{dx_b} \qquad \phi_{2b} = -\frac{\delta d\phi_{2b}}{dx_b}$$

with the resulting core equation expressed by

$$\left\{\frac{1 - \frac{\nu}{\mu\Delta}\left(\frac{1 + \nu\delta}{1 + \mu\delta \cot \mu a}\right)}{1 - \frac{1}{\Delta}\left(\frac{1 + \nu\delta}{\tan \mu a + \mu\delta}\right)}\right\} = \left\{\frac{1 + \frac{\nu}{\mu\Delta}\left(\frac{1 + \nu\delta}{1 + \mu\delta \cot \mu b}\right)}{1 - \frac{1}{\Delta}\left(\frac{1 + \nu\delta}{\tan \mu b + \mu\delta}\right)}\right\}$$

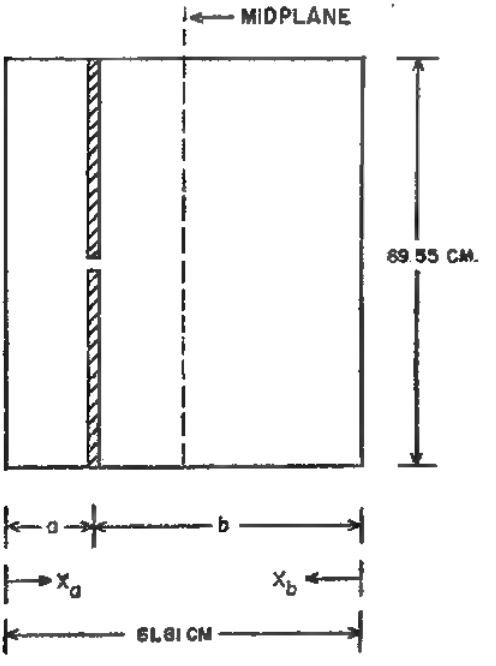

FIG. 3. Schematic diagram of PRR–1 core with two blades inserted.

The control worth of two adjacent blades fully inserted in the core is 6.5 per cent.

TABLE 2.—*Method A. Two adjacent control blades inserted.*

Constants used:

$a = 19.304$ cm	$b = 42.506$ cm
$\tau = 50.7793$ cm^2	$1/\tau = 0.01969$ cm^{-2}
$L^2 = 7.0143$ cm^2	$\delta = 0.784$ cm
$\alpha^2 + \beta^2 = 0.00367$ cm^{-2}	$B^2 = 0.00625$ cm^{-2}
$k_{rodded} = 1.4648$	$k_{unrodded} = 1.3751$

Worth of two adjacent control blades: 6.52 per cent

k	B_1^2	B_1^2	μ^2	ν^2	L	R
1.460	0.00760	−0.16986	0.00394	0.17353	5.8094	6.6364
1.465	0.00768	−0.16994	0.00401	0.17361	5.8432	5.8153
1.470	0.00776	−0.17002	0.00409	0.17369	5.8893	5.1060

METHOD B

In the calculation of the control worths using the second method the formulas of Deutsch(4) were utilized. Since the neutrons are divided into three energy groups, new core constants were calculated on the basis of Deutsch's(5) formulation. These energy groups are:

Fast group	10 Mev to 180 kev
Epithermal group	180 kev to E_c
Thermal group	E_c to 0

where E_c is the thermal cut-off.

a. *Thermal group.*—An effective neutron temperature T_{eff} for the thermal neutrons was calculated from

$$T_{eff} = T\left(1 + C\frac{\Sigma_a}{\xi\Sigma_s}\right)^2$$

where T = temperature of the core = 298°K.

Σ_a = macroscopic absorption cross section at T
Σ_s = macroscopic slowing-down cross section just above E_c
C = 0.89 (obtained from Brown(6) by interpolation)
$\Sigma_a/\xi\Sigma_s = 0.1081$

The atom densities used were taken from Juliano (3) where a T_{eff} of 86°C was obtained using the SOFOCATE code. The value of T_{eff} for this report is 85°C, a very close agreement with the machine-calculated value of 86°C.

Table 3 gives the PRR–I cross sections at 2200 m/sec which were used to calculate $\frac{\Sigma_a}{\xi\Sigma_s}$.

TABLE 3.—*Cross sections at 2200 m/sec.*

Atom	σ_a^*	Σ_a	σ_s	Σ_s
H-1	0.33 barns	0.01237203 cm^{-1}	20.0 barns	0.74982 cm^{-1}
Al-27	0.23	0.00596827	1.4±0.1	0.00261
O-16	0.0	0.0	4.2±0.3	0.00345
U-235	694±8	0.06302908	10±2	0.00001
U-238	2.71±0.02	0.009702342	10±2	0.00003
Total		0.0823396 cm^{-1}		0.7691 cm^{-1}

* Hughes, D. J., and R. B. Schwartz. Neutron Cross Section. Brookhaven National Laboratory Report BNL-325 (2nd edition) (1958).

The value of the thermal cut-off energy, E_c, was determined using neutron conservation between energy groups, i.e.,

$$(E_c/T_{eff})^3\, e^{-(E_c/T_{eff})} = \frac{\Sigma_a}{\xi\Sigma_s}$$

which gives a value of $E_c = 0.1764$ ev. or $E_c = 5.108\ T_{eff}$.

The calculated cross sections for an effective temperature of 86°C are listed in Table 4 below.

TABLE 4.—*Cross sections at T_{eff} of 85°C.*

Atom	$\sigma_{s\ avg}$	$\sigma_{a\ avg}$	$\sigma_{tr\ avg}$	$\Sigma_{tr\ avg}$
	barns	barns	barns	
H-1	29 50439	0.2615774	29.7689674	1.1160684 cm^{-1}
Al-27	1 240722	0.18440243	1.4251244	0.0369806
O-16	3.456297	0.0	3 456297	0.0647917
U 235	9.9716312	534.15772	544 12985	0.0494178
U-238	9.9719888	2.1737417	12.1447303	0.004348
Total				1.2716 cm^{-1}

Summary of Group 3 (thermal)

Energy Range: 0 – 0.1764 ev

$D_3 = 0.262136$ cm

$\Sigma_{a3} = 0.070995384\ cm^{-1}$

$\Sigma_{tr\ avg\ 3} = 1.27160657\ cm^{-1}$

$\Sigma_{f3} = 0.040973142\ cm^{-1}$

$L_3^2 = 3.6922906\ cm^2$

$\Sigma_{aU3} = 0.0485122\ cm^{-1}$

b. *Epithermal group.*—The neutron age, τ_2, was determined by the method of equivalence factors which states

$$\tau_2 = \frac{\left[6.9 + 0.44(u_c - 15.75)\right](1 + V_{Al})^2}{(1 + V_{Al}\,Y_{2Al})\,(1 + V_{Al}X_{2Al})} \qquad (21)$$

where $u_c = 17.8563$ (lethargy equivalent to 0.1764 ev.)

V_{Al} = ratio of the volume of aluminum to the volume of water in the core = 0.77031

$Y_{2Al} = 0.006$

$X_{2Al} = 0.195$

The effect of uranium on the age calculation was neglected in these calculation with the following results:

$\tau_2^{(1}$ (from 180 kev to 1.44 ev, the indium resonance) = 18.71 cm^2

$\Delta\tau$ (from 1.44 ev to 0.1764 ev) ---------- 2.51

τ_2 (from 180 kev to 0.1764 ev) ---------- 21.22 cm^2

SUMMARY OF GROUP 2 (EPITHERMAL)

Energy range: 0.1764 ev - 180 kev

$$D_2 = \frac{1}{3\Sigma_{tr\ 2}} = 0.94063643 \text{ cm.}$$

$$\Sigma_{a2} = \frac{4}{\sqrt{\pi}} (T_{eff}/E_c)^{1/2} \frac{\Sigma_{a3}}{\Delta u} = 0.00483976 \text{ cm}^{-1}$$

$$\Sigma_{tr\ avg\ 2} = 0.35437 \text{ cm}^{-1}$$

$$\Sigma_{f2} = \frac{4}{\sqrt{\pi}} (T_{eff}/E_c)^{1/2} \frac{\Sigma u3}{1.35\ \Delta u} = 0.0024497 \text{ cm}^{-1}$$

$$L_2^2 = \frac{D_2}{\Sigma_{s12} + \Sigma_{a2}} = 19.131163 \text{ cm}^2$$

$$\Sigma_{s12} = 0.044328 \text{ cm}^{-1}$$

Δu = 13.8563 lethargy units

τ_2 = 21.22 cm^2

c. Fast group.—The fast group age τ_1 was calculated using the method of equivalence factors, namely:

$$\tau_1 = \frac{24\ (1 + V_{Al} + V_U)^2}{(1 + V_{Al}Y_{1Al} + V_UY_{1U})(1 + V_{Al}X_{1Al} + V_UX_{1U})} \qquad (22)$$

where $Y_{1Al} = 0.103$ $\qquad X_{1Al} = 0.922$

$Y_{1U} = -0.13$ $\qquad X_{1U} = 1.1214$

giving τ_1 = 41.15 cm^2

SUMMARY OF GROUP 1 (fast)

Energy range: 180 kev to 10 Mev

$$\Sigma_{tr\ 1} = 0.19296\ cm^{-1}$$

$$D_1 = \frac{1}{3\Sigma_{tr1}} = 1.72747\ cm$$

$$\Sigma_{s1\ 1} = \frac{D_1}{\tau_1} = 0.04198\ cm^{-1}$$

$$\tau_1 = 41.15\ cm^2$$

d. *Control worths of slab-type bank of control blades.*—This method is based on dividing the neutrons into three energy groups and that thermal and epithermal neutrons are affected by the control blades while fast neutrons do not see the presence of the control blades. Furthermore, it is assumed that the poison of the blades is uniformly distributed in the core. The k_{eff} of the core is given by (4)—

$$k_{eff} = \frac{\nu \left(F_2 \frac{\Sigma_{f2}}{\Sigma_{a2}} + \frac{F_2 F_3}{1 + B^2 L_3^2} \frac{\Sigma_{f3}}{\Sigma_{a3}} \right)}{(1 + B^2 L_1^2)(1 + B^2 L_2^2)\left[1 + (\Sigma_{a2}/\Sigma_{m2})\right]} \qquad (23)$$

where $F = (A_c - A_R)/A_c$

A_c = physical area of the core cross-section which was 4298.8855 cm² when a reflector savings of 11.57 cm for each graphite reflected side was used.

A_R = absorption area for the control blades

$$A_{2R} = \frac{4aL_2}{1 + (d_2/L_2)} \left[1 + \frac{1}{1 + (d_2/L_2)} \frac{L_2}{2a} \right] \qquad (24)$$

$$A_{3R} = \frac{4aL_3}{1 + (d_3/L_3)} \left(1 - \frac{1}{\left[1 + (L_3/L_2)\right]\left[1 + (d_2/L_2)\right]} \right) \left(1 + \frac{1}{\left[1 + (d_3/L_3)\right]} \frac{L_3}{2a} \right) \qquad (25)$$

a = half-span of the slab = 23.205 cm

B^2 = buckling = 7×10^{-3} cm^{-2}

d = extrapolated distance of epithermal and thermal groups into the blade = $\frac{2\Sigma_{tr}}{3}(1 + P)(1 - P)$

P = transmission probability

The results for the control-blade worths in the PRR–I with the assumptions used are given in Table 5.

TABLE 5.—*Method B. Control worths of all four blades inserted in PRR–I.*

Assumptions	F_3	F_4	Control blade worths $(\frac{\Delta k}{k})$ *Per cent*
Blades black to thermal and epithermal neutrons	0.8592	0.9654	16.79
Blades black to thermal and gray to epithermal neutrons			
a. P = 0.41 (PRR–I condition)	0.9025	0.9654	12.59
b. P = 0.50	0.9141	0.9654	10.70
Blades black to thermal and transparent to epithermal neutrons	1.0	0.9654	3.15
Blades transparent to thermal and black to epithermal neutrons	0.8592	1.0	14.08

CONCLUSION

It is the conclusion that the control and safety blades of PRR–I are sufficient to shut the reactor in the event of a sudden increase in k_{excess} in the core. However, no definite answer as to which of the two methods used is correct can be given until the experimental values are available.

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THE USE OF LOCALLY MADE PLASTER OF PARIS AS DENTAL AND ORTHOPEDIC MATERIAL

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Plaster of Paris, which owes its name to the fact that it was first derived from gypsum obtained at Montmartre, near Paris, is an incompletely hydrated calcium sulfate having the formula $2CaSO_4.H_2O$ (more commonly written $CaSO_4.\frac{1}{2}H_2O$). It is obtained by heating gypsum or plaster stone, a naturally occurring mineral.

In the Philippines, gypsum rocks are available in areas like Batangas, Mindoro, and the Bicol Regions. Erfe, et al.(3) studied the manufacture of plaster of Paris from such materials obtained from Nabua, Camarines Sur. They showed that data obtained from their experiments proved satisfactory compared with data obtained from imported plaster.

Previous studies on the use and application of plaster casts have been made. A. Matthysen, a medical officer in the Dutch Army, invented and described his plaster of Paris bandage in 1852. He employed a coarse cotton bandage with plaster of Paris rubbed into it. Captain J. Vernon Luck,(5) of the United States Army, discussed an improved technique in the application of casts. According to him, unsatisfactory casts are not due to defective plaster but due to poor technique.

The purpose of this study, the experiments of which were done on some employees and patients of the medical and dental clinic of the National Institute of Science and Technology, is to find out if local plaster materials could be used for dental and orthopedic purposes in place of imported plaster.

EXPERIMENTAL PROCEDURE

The plaster of Paris used in this study, the materials for which came from the Ceramic Section of the National Institute of Science and Technology, was made by grinding local gypsum rocks, passing the ground material through a fine sieve, and heating it until it appeared to "boil" in the kettle. It was stirred constantly with a wooden stick until it reached a temperature of

about 170 to 180°, which was maintained for 4 hours to remove half of the combined water present; then it was cooled and sifted.

The plaster was mixed with a suitable amount of water so as to form a paste and allowed to stand. It hardened into a kind of white porous stone which, after careful drying, became unaffected by moisture.

Different plaster-to-water ratios were tried to find out what proportion would yield a good setting, which is the solidification or hardening of the mixture. Good plasters usually set in not less than 10 minutes. Binding agents were also employed to find out which additive was best to obtain the properties of a good orthopedic plaster.

RESULTS AND DISCUSSION

Table 1 shows the different consistencies of plaster that have been tried together with the different setting time.

TABLE 1.—*Characteristics of different plaster consistencies.*

Volume of water (Per 100 gms)	Setting time	Setting temperature	Specific gravity
cc	*Minutes*		
70	18	34.2	1.219
65	20	34.1	1.288
60	21	34.6	1.351
With 5 per cent kaolin			
cc			
70	26	36	1.074
65	26	37	1.095
60	26	38	1.780

As shown in Table 2, plaster bandage made from calcined gypsum with 5 per cent alum and 5 per cent kaolin set satisfactorily in 10 minutes at 32°C just like imported plaster. The only difference is in color, which is not as white as imported plaster.

The above plaster mixture was applied to dental work by using the following water-powder combinations:

Mixture I—Ratio of powder and water is 1:1. The average setting time was 8 minutes. Mixture II—Ratio of powder and water was 1:2. The average setting time was 10 minutes. Mixture III—Ratio of powder and water was 1:3. The average setting time was 15 minutes.

Dental impressions taken from patients showed that, of the three mixture, Mixture III gave the best results. It sets at 15 minutes and hardened completely in 24 hours.

TABLE 2.—*Results obtained in mixing local gypsum with different binding agents in varying proportions.*

Mixture	Percentage composition	Setting time	Setting temperature	Results
	Per cent	*Minutes*	°C	
Plaster	88.0			
Kaolin	10.0	----------	35.0	did not set
Acacia	1.0			
Benzoic acid	1.0			
Plaster	95.0			
Boric acid	5.0	20	----------	Weak
Plaster	95.0			
$CaCO_3$	5.0	----------	-----	did not set
Plaster	93.02			
Kaolin	4.65	----------	----------	Setting uneven
$ZnSO_4$	2.33			
Plaster	92.78			
Kaolin	5.16	11	36.0	Hardened after 3 days with minimum stirring.
$ZnSO_4$	2.06			
Plaster	86.0			
Kaolin	10.0	18	32.0	Weak for orthopedic use
$ZnSO_4$	4.0			
Plaster	98.0			
Alum	2.0	10	34.0	Sufficiently strong for orthopedic use
Plaster	90.0			
Alum	5.0			
Kaolin	5.0	10	32.5	Strong: not as white as imported plaster

SUMMARY

Locally made plaster of Paris was tried for its possible use as dental and orthopedic material in place of imported plaster. After many trials, the material was found to produce encouraging results. These results show local manufacturers their chances of success in making plaster of Paris from gypsum, the utilization of which is certain to bring some good to the country's economy.

ACKNOWLEDGMENT

The authors wish to express their gratitude to Dr. Clodualdo S. Angbengco of the Dental Clinic, National Institute of Science and Technology, for clinically testing our local plaster, and to Mr. Carmelo Erfe, Chief of the Ceramic Section of the same Institute, for his technical assistance.

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THE NUCLEIC ACID OF EHRLICH ASCITES CARCINOMA CELLS, II

ISOTOPE LABELLING

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SEVEN TEXT FIGURES

Attempts have been made by several investigators to clarify the relationships existing between the nucleic acids present in the different subcellular structures. Many of their investigations were effected through a study of the specific activities of the RNA and DNA isolated from these fractions at different times after the administration of an isotopically labelled precursor. Thus, time-activity studies carried out by Potter,(8, 10) using C^{14} labelled orotic acid showed that the specific activity of the nuclear RNA increased much more rapidly and reached a higher level than did that of the cytoplasmic RNA. Similar results were obtained by Tyner, et al.(13) who used glycine-2 C^{14} as the labelled precursor.

It was felt, therefore, that an investigation into the kinetics of P^{32} phosphate incorporation into the nucleic acids of the centrifugally isolated subcellular fractions, complemented by studies of the isotope labelling of the nucleic-acid elution patterns obtained from these fractions by chromatography on ECTEOLA-cellulose, would help in the elucidation of the relationships existing between these cell compartments.

MATERIALS AND METHODS

The centrifugation and chromatographic procedures used in this paper for the isolation of the subcellular nucleic acids have been described previously.(5)

Incorporation of P^{32}.—The Ehrlich ascites carcinoma cells were removed from the animal and then washed free of blood elements by repeated suspension and recentrifugation in isotonic saline. The packed cells were diluted to eight times their volume with ascites fluid previously collected. One ml of such a suspension was added per 3 ml of total volume and also a total of

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approximately 100 uC of P^{32} as inorganic phosphate. Glucose to a concentration of 0.02 *M*, as well as *mM* phosphate buffer, pH 7.4 was added before incubation of the cells at 37°C in a shaking water bath.

With *in vivo* experiments using rat liver, the animal was placed in a restrainer with the tail protruding. Inorganic P^{32} was then injected intravenously via the inferior caudal vein using a hypodermic syringe fitted with a No. 27 needle. The rats were subsequently sacrificed after the prescribed incubation time.

After incubation, the cells were fractionated centrifugally and aliquot samples chemically treated to characterize the incorporated radioactivity. In the chromatographic studies, the centrifugally obtained fractions were absorbed on DCTE-OLA-cellulose and eluted by solutions of NaCl.(5) Radioactivity measurements were then carried out on the eluted samples after estimation of the nucleic-acid content. Samples showing maximal UV absorption and radioactivity were fractionated chemically in order to characterize the radioactive constituents.

Chemical fractionation.—Samples were precipitated with 5 per cent trichloracetic acid (TCA) and cooled for one hour at 4°C. The supernatant after centrifugation was decanted and an aliquot sample plated and counted. This fraction, called the acid-soluble phosphate fraction, was extracted three times with twice its volume of ether to remove TCA and then evaporated to dryness. The material was then subjected to two-dimensional paper chromatography on Whatman No. 1 filter paper using 0.1 *M* sodium phosphate pH 6.8 (100 ml)—ammonium sulfate (60 g)—n, propanol (2 ml) and ethanol (70 ml)—1.0 *M* ammonium acetate pH 7.5 (30 ml) as first- and second-dimension solvents, respectively. Radioactive areas on the chromatogram were located by radioautography using Kodak 'no screen' x-ray film and counted directly on the paper.

The acid-insoluble material was washed twice with 5 ml portions of 5 per cent TCA, and then extracted with ethanol-ether (1 : 1) at 60°C for 30 minutes to remove the lipid material.

The insoluble matarial left from the treatment was dissolved in *N* KOH and left to stand overnight at room temperature. Protein and DNA were precipitated from this solution by bringing the pH to 3.5 with perchloric acid. The precipitate was removed by centrifugation, and a sample of the supernatant fluid, containing the 2', 3'-ribonucleotides derived from RNA and also inorganic phosphate from the hydrolysis of phospho-

protein, was plated and counted. The inorganic and nucleotide phosphates were separated by electrophoresis of the mixture at pH 3.5 in 0.05 *M* ammonium formate,(4) the different bands located by radio-autography and their activities counted on paper. The precipitate resulting from the addtion of perchloric acid was washed with cold 5 per cent TCA, resuspended in water, plated and counted. This is referred to as the DNA fraction.

RESULTS

An investigation of the kinetics of P^{32} phosphate incorporation into the nucleic acids of the cell nucleus was initially carried out. In the experiments quoted here, the intact ascites cells were disrupted by high-speed homogenization in a sucrose-calcium medium. The nuclei were then separated from cell debris by means of differential centrifugation. The isolated nuclei, upon microscopic examination, were observed to be intact and free from disintegrated cell particles.

Incorporation of P^{32} into nuclear components.—Two procedures were available for studies on P^{32} incorporation into the nucleic acids present in the cell nucleus. One of these was the prior isolation of nuclei from the cell and their subsequent incubation with P^{32} in an appropriate medium. The other procedure involved the incubation of the intact cell under the prescribed experimental conditions, after which the nuclei were isolated and isotope incorporation into the nuclear components studied.

The first of these procedures was found to be unsatisfactory as the isolated nuclei coagulated in the incubation medium even after the addition of various anticoagulants. The second proceddure was therefore adopted for the experiments reported here. Ascites cells were incubated with P^{32} in ascitic fluid, samples were then taken at various times and homogenized. The separated nuclei were precipitated with 5 per cent TCA and fractionated chemically to characterize the incorporated radioactivity. The results obtained are detailed in Table 1. It may be seen that the nucleic acids present in the cell nucleus incorporated radioactivity from the P^{32} labelled phosphate and that this incorporated radioactivity increased with time for periods up to 3 hours. Uptake of P^{32} into the acid-soluble, low-molecular-weight components of the nucleus on the other hand, increased rapidly in the first hour but subsequently declined. This result suggests a precursor-product relationship

between the transient intermediates present in the acid-soluble fraction and the nucleic acids in the acid-insoluble fraction.

TABLE 1.—*Time course of P^{32} incorporation into Ehrlich ascites nuclear components.*

Time (min)	Mumoles P^{32} Incorporated and cells		
	RNA	DNA	Acid solubles
30	84	22	726
60	193	57	955
120	276	132	725
180	467	201	709

Whole cells incubated at 37°C in ascitic fluid with 100 uC P^{32} for the required times.

P^{32} incorporation into RNA of the subcellular fractions.—It has been previously observed that isotope incorporation into the nuclear ribosenucleic acids (nRNA) occurs at a much higher rate when compared to that of the cytoplasmic RNA (cRNA) fractions. This observation has been consequently interpreted to mean that nRNA is the precursor of cRNA.(9) A time study of the incorporation of P^{32} into the RNA of the subcellular fractions of Ehrlich ascites cells was therefore undertaken to determine the relationships existing between the nuclear and cytoplasmic fractions, if any. In this experiment, the ascites cells were incubated in ascitic fluid with P^{32} and samples taken after certain periods. The cells were washed, homogenized, and the subcellular fractions obtained as described previously. The RNA nucleotides from these fractions were subsequently isolated by chemical fractionation procedures and their specific activity determined. The results showed that the specific activity of nuclear RNA from Ehrlich ascites cells occurred earlier and increased at a greater rate as compared to the RNA from any of the cytoplasmic fractions. This is clearly illustrated in Fig. 1. The increase in activity of nuclear RNA continued at the initial high rate for periods of up to 6 hours. The incorporation of P^{32} into microsomal RNA was observed to proceed after an initial lag period of 90 to 120 minutes, but thereafter increased at a greater rate. The time course of P^{32} incorporation into the post-microsomal RNA and supernatant RNA was observed to be essentially linear, no lag period being exhibited in either case.

In order to gain a clearer insight into the incorporation patterns of P^{32} and in particular, to determine the relative stability and turnover of incorporated radioactivity in the different

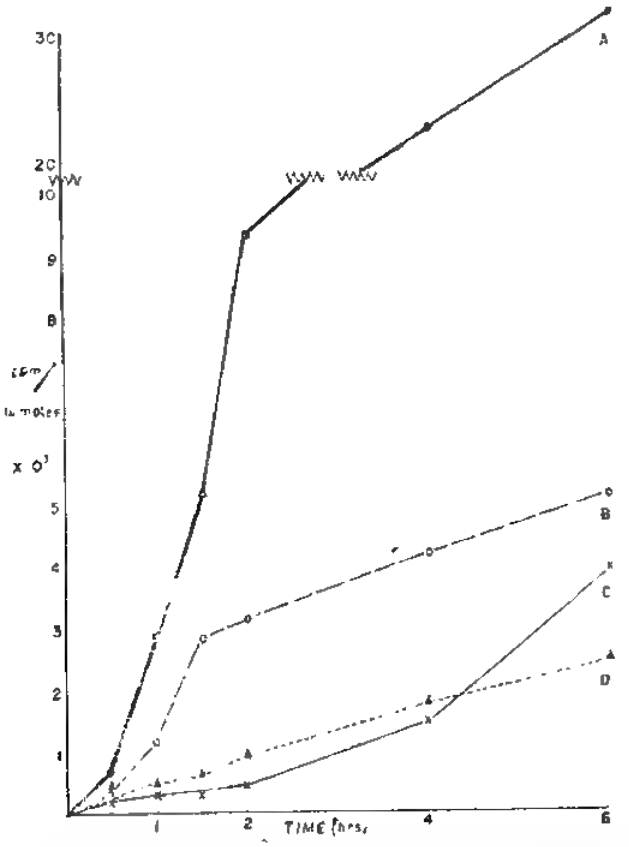

FIG. 1. The course of labeling with P^{32} of the subcellular fractions of Ehrlich ascites cells. Incubation was at 37°C in ascitic fluid for the times indicated. Samples were then removed and fractionated centrifugally as described in the text. RNA nucleotides of each centrifugal fraction were isolated and their radioactivity determined. Activity expressed as counts/minutes per m_μ mole of RNA.

A, Nuclear RNA; *B*, Supernatant RNA; *C*, Microsomal RNA; *D*, Post-microsomal RNA.

fractions, a "pulse" experiment was carried out. This type of experiment, pioneered by Bolton, et al.,(3) makes it possible to gain information concerning the pathways of phosphate metabolism by incubating cells for a certain period of time in a medium containing radioactive phosphate and then following the redistribution of the isotope after replacement of the radioactive phosphate in the medium by non-radioactive phosphate. This procedure has been applied to the study of P^{32} incorporation in Ehrlich ascites cells and was effected by incubation with the

isotope for 30 minutes, centrifugation, then resuspension in fresh medium containing no isotope. Samples were taken at regular time intervals, the cells disrupted, and the subcellular fractions obtained by differential centrifugation as described previously. The RNA nucleotides were subsequently isolated and their specific activities determined.

The results shown in Fig. 2 indicated that the radioactivity incorporated into the RNA of the nuclear fraction continued to increase for at least 90 minutes after removal of the isotopic phos-

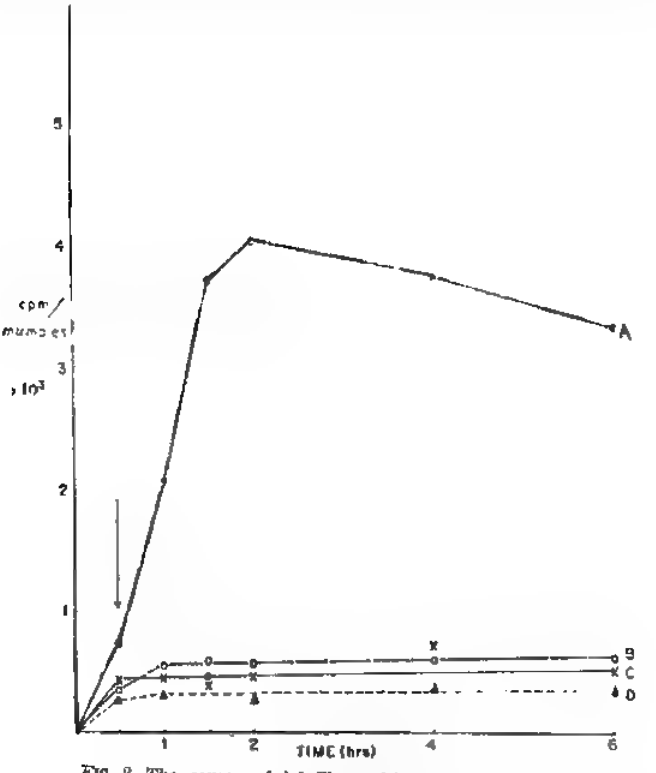

FIG. 2. The course of labelling with P^{32} of the subcellular fractions of Ehrlich ascites cells after replacement of P^{32} in the ascitic fluid medium by nonradioactive P^{32} phosphate. Replacement was carried out after 30 minutes of incubation. Samples were removed at indicated times and fractionated centrifugally. RNA nucleotides of each fraction were isolated and the radioactivity assayed. Activity expressed as count/minute per mμ mole RNA.

A, Nuclear RNA; *B*, Supernatant RNA; *C*, Microsomal RNA; *D*, Post-microsomal RNA.

phate, after which time it declined. The incorporation of P^{32} into the other cytoplasmic fractions ceased soon after replacement of the radioactive medium, the specific activities remaining

at approximately the same levels for the duration of the experiment quoted. This result would seem to indicate, therefore, that RNA from the nuclear fraction of Ehrlich ascites cells does not act as a precursor for the RNA of the cytoplasmic fractions. However, it is possible that any precursor-product relationships between the nRNA and RNAs present in the cytoplasmic fractions may have been masked by the heterogeneity of the RNA present in these fractions. Evidence has been obtained in a previous paper of the existence of such a heterogeneity in the RNA of the subcellular components and this observation has been corroborated by several other workers.(1, 5, 11, 12) This heterogeneity in RNA has been demonstrated through various procedures, such as analytical ultracentrifugation, ion-exchange chromatography and electrophoresis and, thus, cannot be construed as an artifact of the procedure employed. One such method employing the anion exchanger ECTEOLA-cellulose has been found particularly valuable in our investigations for the isolation and purification of ribonucleic-acid-containing materials.

Isotope labelling of nucleic-acid chromatography patterns.— An effort to correlate the radioisotope labelling of the nucleic acids of the subcellular fractions with the nucleic acid chromatography patterns previously obtained,(5) was attempted in the next series of experiments. It was hoped thereby to determine the effects of heterogeneity in the subcellular nucleic acids on the isotope incorporation picture.

The incubation of Ehrlich ascites carcinoma cells with P^{32} labelled inorganic phosphate for two hours prior to fractionation resulted in a radioactivity pattern shown as an unmarked area in the nucleic-acid chromatogram of the nuclear fraction (Fig. 3). The nucleic-acid areas are shown in the stippled area for comparison purposes. It will be noted that there is a correspondence between the nucleic-acid and radioactivity peaks in the pattern except for the first peak of radioactivity which was eluted by 0.06 *M* NaCl. Chemical fractionation of the radioactive regions showed that the radioactivity in this early peak was mainly associated with inorganic phosphate found in the TCA soluble fractions, while the activity shown by the 0.34 *M* NaCl elution was associated mainly with RNA. Radioactivity incorporated into DNA was observed to account for about 30 per cent of the activity in the fraction eluted by 0.53 *M* NaCl, the remainder of the incorporated radioactivity being distributed as follows: phosphoprotein 36 per cent, RNA 20 per cent, and

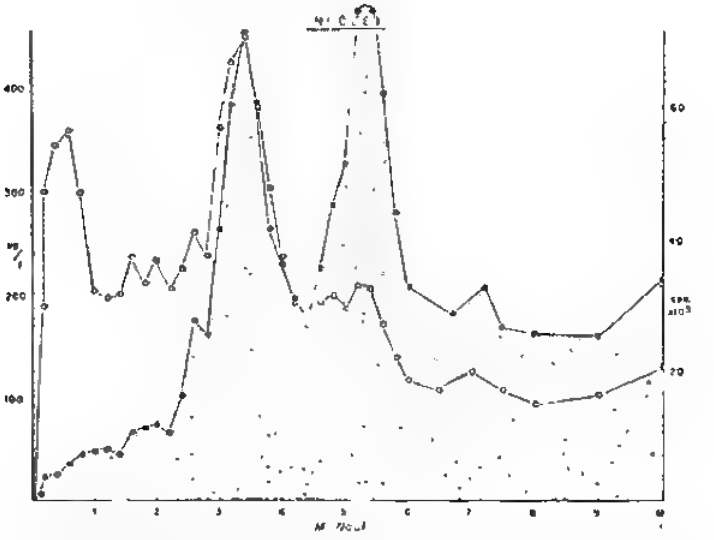

FIG 3. Nucleic-acid and P^{32} labelling patterns of the nuclear fraction of Ehrlich ascites carcinoma cells upon chromatography on LCTEOLA-cellulose anion exchanger. Stippled areas represent nucleic-acid regions expressed as μg of nucleic acid/eluted sample and unmarked areas, radioactive regions expressed as counts per minute of eluted samples. Cells were incubated at 37°C for 2 hours with 0.5 mC of P^{32} in ascitic fluid.

phospholipid 10 per cent. An incubation period of an hour resulted in the radioactive labelling of the fractions eluted by 0.06 and 0.34 *M* Na Cl only.

Only slight incorporation of radioactivity was noted into the different nucleic acid components of the microsomal fraction after an hour of incubation with P^{32} labelled phosphate. After 2 hours of incubation, however, the radioactivity pattern shown as an unmarked area in Fig. 4 was observed. It may be seen that there is again a very close correspondence of the nucleic acid peaks with the radioactive peaks. As with the nuclear fractions, the high radioactivity exhibited by the early elutions was found to be associated principally with the inorganic phosphate in the acid-soluble fraction, while the radioactivity in the later elutions was accounted for mostly by RNA.

Radio-assay of the nucleic-acid chromatogram given by the post-microsomal fraction of Ehrlich ascites cells gave results illustrated in Fig. 5. The radioactivity pattern shown in the unmarked area of the chromatogram was obtained after a 2-hour incubation run with P^{32} labelled phosphate. The major

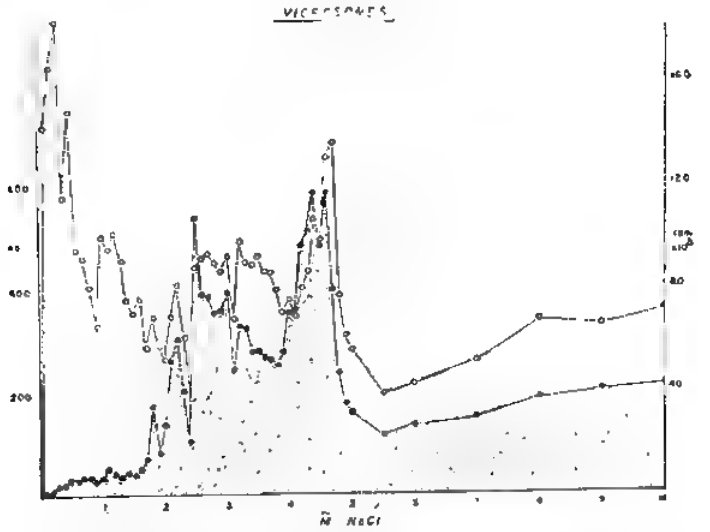

Fig. 4. Nucleic-acid and P^{32} labelling patterns of the microsomal fraction of Ehrlich ascites cells upon chromatography on ECTEOLA-cellulose. Stippled area represent nucleic-acid regions expressed as ug NA/eluted sample and unmarked areas, radioactive regions expressed as count/minute of eluted samples. Conditions of incubation as in Fig. 3.

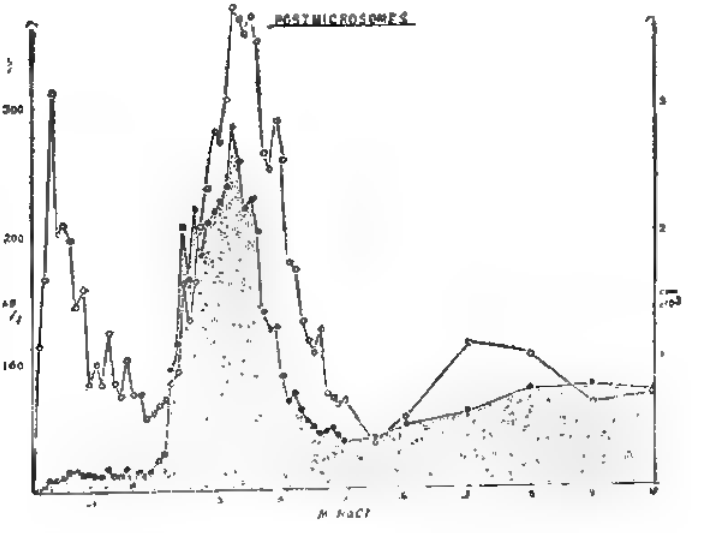

Fig. 5. Nucleic-acid and P^{32} labelling patterns of the postmicrosomal fraction of Ehrlich ascites carcinoma cells upon chromatography on ECTEOLA-cellulose. Stippled areas represent nucleic-acid regions and unmarked areas, radioactive regions. Conditions of incubation as in Fig. 3

component was found to be labelled in toto, the radioactive regions closely paralleling the nucleic-acid areas. As with the two preceding fractions, the high amount of radioactivity shown by the samples eluted with the lowest salt concentration was found to be associated primarily with inorganic phosphate. Radioactivity incorporated in RNA accounted for about 60 to 70 per cent of the activity exhibited by the major component while protein phosphorus accounted for about 25 to 30 per cent of the labelling. Only slight radioactivity was noted to be incorporated into the nucleic-acid component eluted by 0.2 to 0.4 *M* NaCl after one-hour incubation with P^{32} labelled phosphate.

Hoagland, et al.(7) observed that radioactive leucine was bound to RNA found in the supernatant of liver tissue homogenates which had been centrifuged for 2 hours at 105,000 g. This RNA was chromatographed by Goldthwait(6) on ECTEOLA-cellulose and was eluted as two components, one by 0.15 to 0.2 *M* NaCl, and the other by 1.0 *M* NaOH. In our previous experiments, it was observed that the RNA present in the supernatant fraction of Ehrlich ascites cells chromatographed on ECTEOLA-cellulose was such that over half of the nucleic-acid material was eluted by 0.1 to 0.3 *M* NaCl. The radioactivity pattern obtained from this fraction after a 2-hour incubation with P^{32} labelled phosphate is shown in the unmarked areas of Fig. 6. It will be noted that the bulk of the radioactivity incorporated was eluted by 0.01 to 0.09 *M* NaCl. Chemical fractionation procedures showed that this radioactivity was linked mainly to inorganic phosphate and partly with the acid-soluble nucleotides, accounting for about 90 per cent of the total radioactivity incorporated by the cell. It is therefore possible that the high radioactivity eluted by 0.01 to 0.09 *M* NaCl in the other subcellular fractions may be due to contamination by supernatant inorganic phosphate. The RNA component of this fraction which was eluted by 0.2 to 0.3 *M* NaCl was observed to be isotopically labelled as early as one hour after incubation with P^{32}.

For comparison purposes, a study of the P^{32} labelling of the nucleic-acid elution pattern from the subcellular fractions of normal rat-liver tissue was undertaken and the results are shown in Fig. 7. Examination of the activity patern shows close correlation between the maximal UV-absorbing areas and the peak of the radioactivity regions. Chemical fractionation

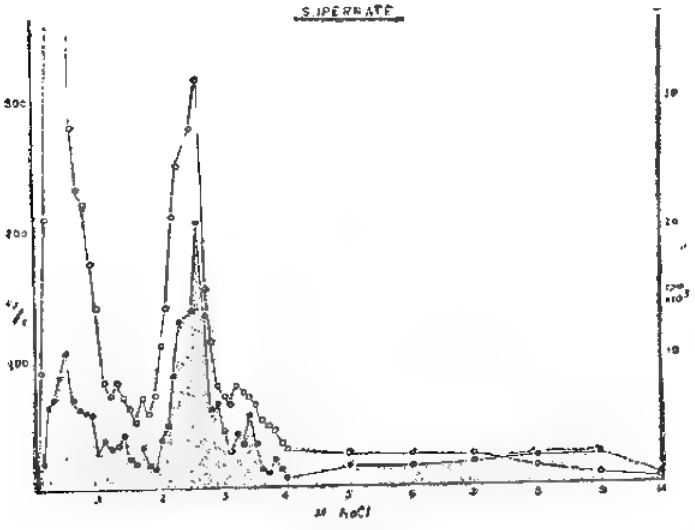

FIG. 6. Nucleic-acid and P^{32} labelling patterns of the supernatant fraction of Ehrlich ascites carcinoma cells. Stippled areas represent nucleic-acid regions and unmarked areas, radioactive regions. Conditions of incubation as in Fig. 3.

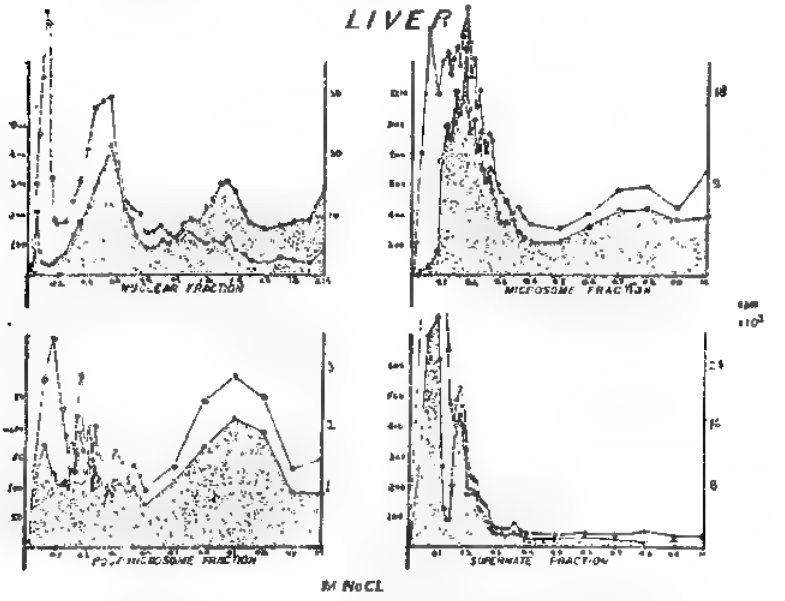

FIG. 7. Nucleic-acid and P^{32} labelling patterns of the subcellular fractions of normal rat-liver cells upon chromatography on ECTEOLA-cellulose. Stippled areas represent nucleic-acid regions and unmarked areas, radioactive regions. Rats were injected with 1 mC of P^{32} labelling phosphate and the liver excised and fractionated 5 hours later.

of the radioactive regions corresponding to areas showing maximal UV absorption, showed that the radioactivity of the fractions eluted by salt concentrations of from 0.1 *M* and up, was associated mainly with RNA, while that of fractions eluted by NaCl solutions lower than 0.1 *M* was associated primarily with inorganic phosphate and acid-soluble organic phosphates. A time study of the incorporation of P^{32} into the nucleic-acid patterns obtained from rat-liver subcellular fractions revealed that the incorporation of the isotope did not occur to a significant extent in any of the cytoplasmic fractions until after 3 hours of *in vivo* incubation. After this period the specific activities of the various nucleic-acid peaks progressively increased during the experimental period of 48 hours. On the other hand, incorporation of P^{32} into the nucleic-acid peak eluted by 0.5 to 0.6 *M* NaCl from the rat-liver nuclear fraction was observed as early as 1 hour after injection of the isotope into the rat. Little incorporated radioactivity was noted in the nucleic-acid peak of the nuclear fraction eluted by 1.2 to 1.4 *M* NaCl even after 24 hours of incubation.

DISCUSSION

The results shown in Table 1 indicate that P^{32} labelled phosphate is incorporated into the constituents of Ehrlich ascites cell nuclei when the intact cells are incubated in the presence of the isotope. The rate of P^{32} incorporation into the nucleic acids of the cell nuclei was observed to increase rapidly after the first hour of incubation in spite of a decline in the radioactivity of the TCA soluble constituents. These data are consistent with a precursor-product relationship between the components of the acid-soluble fraction and the nucleic acids of the cell nucleus.

The possibility of nuclear RNA serving as a precursor of cytoplasmic RNA has been suggested by the observations of several workers that the specific activity of nuclear RNA increased rapidly and rose to a considerably higher level than did the specific activity of RNA in any of the cytoplasmic fractions. Our studies on the kinetics of P^{32} incorporation into the subcellular components of Ehrlich ascites cells have confirmed the observation that the RNA present in the nucleus of the cell exhibited the most rapid and the highest incorporation of the isotope. Moreover, no significant incorporation of radioactivity into the nucleic acids of the cytoplasmic fractions, obtained from both Ehrlich ascites cells and rat-liver tissue, was observed until after 2 to 3 hours of incubation with the isotope. However, an investigation of P^{32} turnover in these fractions did not

provide evidence for the view that nuclear RNA serves as a precursor for cytoplasmic RNA. Tracer experiments conducted by Barnum, et al.(2) have tended to support this latter finding. Nevertheless, our results did not rule out the possibility that any precursor-product relationship may have been obscured by heterogeneity existing in the nucleic acids of the subcellular fractions, inasmuch as these nucleic acids could not be distinguished within the limits of the experimental procedure employed.

The results obtained in our preceding paper did indicate the existence of heterogeneity in the nucleic acids of the subcellular fractions. However, although marked heterogeneity does exist between different nucleic-acid components of the various cytoplasmic fractions, it has only been possible to show a difference in the rate of labelling with P^{32} in the case of nuclei. This was observed in the nuclear fraction of Ehrlich ascites cells, where it was found that the nucleic-acid peak eluted by 0.34 *M* NaCl was labelled earlier than the nucleic-acid peak eluted by 0.53 *M* NaCl. Non-uniform labelling was also observed in the nuclear fraction of rat-liver tissue, where the nucleic-acid peak eluted by 0.5 to 0.6 *M* NaCl was labelled one hour after injection of the isotope into the rat. The incorporation of P^{32} into the cytoplasmic fractions appeared to proceed at a similar rate into all the chromatographically separated components of each fraction.

SUMMARY

Studies on the P^{32} labelling of the subcellular fractions of Ehrlich ascites carcinoma cells revealed that the nuclear fraction was the first cellular component to become labelled. The incorporation into nuclear RNA was observed to be considerably higher than into the other cytoplasmic fractions. Turnover studies on the P^{32} labelling of these fractions showed no conclusive evidence for the view that cytoplasmic RNA originated from nuclear RNA.

The incubation of the cells with P^{32} labelled phosphate for two hours prior to fractionation resulted in a radioactivity pattern which corresponded closely to the nucleic-acid pattern upon chromatography on ECTEOLA-cellulose.

Chemical fractionation of the peaks showing maximal UV absorption and radioactivity incorporation indicated that the radioactivity of the very early elutions (0.01 to 0.09 *M* NaCl) was associated mostly with inorganic phosphate, while the radioactivity shown by the later elutions was associated mainly with RNA.

ACKNOWLEDGMENTS

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AGE RESISTANCE OF RATS, GUINEA PIGS AND CATS TO TRYPANOSOMA EVANSI WITH A NOTE ON THE BIOMETRY OF THE FLAGELLATE *

By Lope M. Yutuc

Few studies have been conducted relative to the resistance of young and old animals to various species of trypanosome infection. Kligler and Rabinowitch(6) found that on exposure to surra trypanosome infection, rats weighing from 35 to 40 grams died sooner than those weighing from 50 to 60 grams. With *Trypanosoma rhodesiense*, however, Corson(2) recorded little difference in longevity between 10 rats of 50-gram average weight and 10 rats of 100-gram average weight. Kolodny(7) reported that in rats infected with *Trypanosoma cruzi*, a heavy rate of parasitemia was observed and more fatalities occurred in animals less than 20 days old than in older subjects. In a series of experiments, Duca(5) noted that *Trypanosoma lewisi* was more often fatal in young rats than in older ones. Poindexter, cited in Culbertson's paper on the natural resistance against animal parasites acquired as hosts mature, brought out the fact that in *Trypanosoma equiperdum*, younger rats survive the infection for somewhat longer periods than the older animals. This statement was corroborated by Culbertson.(3) Again, Culbertson and Kessler(4) recorded that *Trypanosoma cruzi* was generally more fatal in mice below 25 days of age than in mice above this age, the latter usually surviving the infection. On the other hand, Ashcroft(1) demonstrated that rats weighing 50 grams over infected with *Trypanosoma brucei* lived longer than older and heavier rats when the strain of the infecting trypanosome was maintained in adult rats for more than three months. Conversely, when the infecting trypanosome was maintained in young rats, young rats exposed to this trypanosome lived shorter than older subjects, indicating the plasticity of this strain of trypanosomes.

In the present study, the author reports on the relative age resistance of white rats, guinea pigs and cats to *Trypanosoma evansi* and the biometry of this trypanosome.

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MATERIALS AND METHODS

The strain of trypanosome was isolated from a natural case of equine surra and maintained continuously in guinea pigs by syringe passage for a period of one year, before using it for infecting white rats and guinea pigs in this experiment. In the cat series, the trypanosome used was obtained from the Bureau of Animal Industry and kept in white rats for a period of three months before it was used in Experiment III. Thence, it was maintained through syringe passage from cat to cat until the termination of the study. The animals used in these experiments were laboratory bred. The number of the organisms used per dose and the density of the trypanosome in the blood stream were estimated with the aid of a hemacytometer. The infective dose was the same irrespective of the age group of the host. In other words, young and old subjects were infected with the same number of trypanosomes.

Measurement of the trypanosomes on stained preparations was done with a camera lucida. Microscopic examination of the blood of the experimental subjects was carried daily in the rats and every other day in the guinea pigs and the cats.

RESULTS AND DISCUSSIONS

The results are summarized in Tables 1 and 2. As to the prepatent period of the parasite and the longevity of the host, there is hardly any difference between the young and old rats. While this finding does not support that of Kligler and Rabinowitch with *Trypanosoma evansi* in rats, paradoxically it is in agreement with that of Corson who, using *T. rhodesiense* states that there is little difference in longevity between 10 rats of 50 grams and 10 rats of 100 grams body weight. It is interesting to note that the younger subjects had significantly heavier densities of trypanosomes in the blood at about the terminal course of the infection than the older ones, the range being from 700,000 to 7,000,000 with an average of 5,000,000 in the young rats and 650,000 to 5,000,000 with an average of 2,000,000 in the adults. This observation corroborates that of Kolodny, Culbertson and Kessler in *Trypanosoma cruzi* infection in rats and mice, although the infection in the latter was not as heavy. If this capacity of young rats to sustain a heavier burden of trypanosomes during the agonal stage of the infection but with the same survival period as of the old rats, can be taken to mean an expression of resistance, then the younger rats are

TABLE 1.—*The relative longevity of rats, guinea pigs and cats of different age groups to Trypanosoma evansi.*

Experiment number	Number of animals	Average age in days	Ineffective dose in thousands	Average prepatent period	Length of life in days	Average length of life in days
			WHITE RATS			
I	10	24	5 to 4,000	6.30	6, 10, 12, 2-13, and 14	11.2
	10	836		6.50	5-10, 2-11, and 3-13	11.1
			GUINEA PIGS			
II	11	16	0.75 to 3,000	7.20	12, 14, 18, 23, 26, 48, 59, 66, 72, 113, and 152	52.8
	11	214		14.63	14, 25, 55, 75, 79, 85, 91, 104, 105, 106, and 154	81.2
			CATS			
III	7	33	100	5.3	9, 2-11 2-14, 2-25	15.9
	7	428		6.7	10, 37, 54, 60, 82, 90, and 179	73.5

TABLE 2.—*The length of Trypanosoma evansi in relation to its density in the blood.*

Number of days after positive	Mean length in microns	Coefficient of variation in per cent	Number per cmm
	WHITE RAT		
6th	27.66±0.2410	5.03	12,850
7th	26.06±0.1605	11.51	589,750
8th	24.14±0.2640	10.89	4,961,580
9th	22.56±0.2510	11.36	5,972,040
	GUINEA PIG		
18th	28.37±0.3980	10.93	18,870
20th	26.30±0.8280	1...8	138,040
22nd	24.81±0.7300	13. 0	259,740
28th	24.82±0.4700	11.63	92,240
37th	25.99±0.3800	16.08	17,170

somewhat more resistant than the older. On this score, the above finding tends to give an indirect support to the statement of Poindexter that young rats infected with *Trypanosoma equiperdum* lived somewhat longer than the old rats. Irrespective of the age group, the highest density occurs at about the time of death. Moreover, the increase in the density of the trypanosome while progressive is not geometrical (Table 2).

In general, despite the wide range in the individual resistance of the guinea pigs, there is a decided difference in the prepatent and the longevity periods in favor of old animals in this species. As may be seen in Table 1, Experiment II, the average prepatent period is 7.2 days and the average longevity period is 52.8 days for guinea pigs with an average age of 16 days, while in the 214-day-age group the figures are 14.63 days and 81.2 days, respectively. While parasitemia in the young subjects falls within the range of those observed in the rat group, in most of the old and some of the young guinea pigs, the density of the trypanosome in blood suffers a series of crises and relapses. In the majority of the old and in but few of the young animals, the parasite at about the time of death, could hardly be detected microscopically. The heaviest density noted in the young guinea pigs is 790,000 trypanosomes per cmm while with the old age group, 260,000 trypanosomes per cmm of blood. The latter figures is indicative of the capacity of the old subjects to restrict the multiplication of the parasite better than those of the youngs.

The cat group presents a picture almost intermediate between the rat and the guinea pig. In the kitten group as shown in Table 1, Experiment III, the prepatent period and the duration of the infection are 5.3 and 15.9 days, respectively, which figures are closer to the rat group than to the guinea pig series. However, when the prepatent and the longevity periods which are 6.7 and 73.5 days, respectively, of the adult group are considered, the former figure is almost identical with those of rats of both age groups and of the group young guinea pigs, but the latter figure is very close to the longevity period, of the adult guinea pigs. Likewise, Parasitemia in this group presents an intermediate pattern. In the kitten, the course of the disease is progressive but the trypanosome density of (600,000 per cmm) is not as heavy as that of the rat group, although it falls below that of the young-guinea pig group. In adult cats the 300,000 per cmm density is almost

the same as that of the old guinea pig group, but the trypanosomes in most cases remain in the peripheral circulation until death of the host.

To a certain extent, the present findings lend support to the general statement that the longevity of infected animals of different ages depends not only on the species of animals and species of trypanosomes involved but also on the previous history of the strain of the trypanosome (Ashcroft).

In the horse, Yakimoff, et al.(9) found *Trypanosoma evansi* to be 21.97 microns in length one day and 22.46 microns the day following. Working with *Trypanosoma brucei*, Plimmer and Bradford(8) considered the parasite to vary in size and length with the period of the disease and the species of the host animal. They noted it to be longest in the rat at the time of death.

To test the above statements, observations were made on the length of *Trypanosoma evansi* in relation to the density of the trypanosomes in the circulating blood of rats and guinea pigs. These animals were chosen, because they represented a susceptible species on the one hand and a resistant one on the other. The results may be read in Table 2. In the white rat there was a decided progressive decrease in the length of the flagellate, starting with 27.66 $\pm$ 0.2410 microns in length at a density of 12,850 per cubic millimeter of blood, six days after infection and ending with 22.96 $\pm$ 0.2510 microns in length at a density of 5,972,000 trypanosomes per cubic millimeter of blood shortly before death.

In the guinea pig which became positive on the eighteenth day, the organism measured 28.35 microns $\pm$ 0.3980 at a density of 18,370 trypanosomes in one cubic millimeter of blood. Two days later the size decreased to 26.30 microns $\pm$ 0.8280 at density of 138,650 trypanosomes. When the trypanosome density reached its peak of 259,740 parasites per cubic millimeter of blood, the size decreased to its minimum level of 21.91 $\pm$ 0.330 microns. However, when the density receded to 92,240 organisms per one cubic millimeter of blood, the length rose to 24.83 $\pm$ 0.270 microns on the twenty-eight day. At the last reading, made on the thirty-seventh day, the length further increased to 25.99 $\pm$ 0.380 microns despite a decrease in density in the blood to 17,450 organisms in one cubic millimeter. A return to the original 28.35 $\pm$ 0.3980 microns or thereabout was not attained even when the parasite density was reduced

to a degree lower than the observed when the parasites first measured in blood.

The results presented have shown that the greater the density of the flagellates in the blood the shorter it becomes in length. Another way of expressing it, the faster the organisms divide, the smaller the size. This results support the finding that not only the length varies from day to day in the same host but also with the stage of the disease and the species of the host (Yakimoff, et al. and Plimmer and Bradford).

The day before the rat was found dead, the length of the trypanosome was decidedly shorter than its initial measurement. This finding does not fit in with the statement of Plimmer and Bradford with *Trypanosoma brucei* that "the largest in rat at the time of death." The failure of the trypanosome to reach the original size in the initial reading in the guinea pig in spite of the fact that the density returned to almost the initial level and if this circumstance may find general acceptance, the suggestion it offers, directs one to a developing resistance of the host expressed in the form of reduced length of the trypanosome even when the density in the blood is low. In superinfection in nematodes, stunted growth in the parasite is generally accepted as an expression of increased resistance of the host.

SUMMARY

In the rat, age difference did not show any influence on the course of *Trypanosoma evansi* infection. Young and old subjects succumbed to the disease after almost the same length of exposure. However, the trypanosome density was much greater in the young than in the old even shortly before death supervined. In the guinea pig and the cat, on the other hand, marked age difference in resistance occurred between age groups in favor of the old animals. A majority of the old guinea pigs but only a few of the youngs died with the trypanosomes not demonstrable in the peripheral circulation. The cat appeared to be intermmediate in its response. While the trypanosomes showed hardly any fluctuations in density throughout the infection even up to the time of death of the young hosts, in the old cats as in the old guinea pigs, the parasites fluctuated in number to a much greater degree and very seldom if ever disappeared from the blood even before the death of the host.

In the rat, the flagellates progressively decreased in size during the course of the infection and until death of the victim. The size had a negative correlation with parasite density in the blood. However, in the resistant subject as in the guinea pig, the length of the parasite while generally in inverse proportion to parasite density, fluctuated during the course of the infection and did not return to the original even when the parasite density became lower than that observed when trypanosomes first measured in blood.

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CONTRIBUTIONS TO A REVIEW OF PHILIPPINE SNAKES, II

THE SNAKES OF THE GENERA LIOPELTIS AND SIBYNOPHIS

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The Philippine snakes of two colubrid genera, *Liopeltis* and *Sibynophis*, are treated in this second contribution to a review of Philippine snakes. These genera have restricted distributions in those islands, both being limited to the islands of the Palawan Archipelago. Both, too, are evidently fairly recent derivatives of closely related populations now extant in Borneo.

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TERMINOLOGY

Standard length: distance from tip of snout to anal opening.

*: Following locality under "Range" denotes locality from which material was examined.

Diagnosis: The salient characteristics of the species are summarized under this heading. The measurements given are for the largest specimen, one male and one female, examined.

Genus LIOPELTIS Fitzinger

Liopeltis Fitzinger (1843) 26 (type species *Herpetodryas tricolor* Schlegel, by monotypy).

Phragmitophis Günther (1862) 126 (type species *Herpetodryas tricolor* Schlegel, by monotypy).

Definition.—Maxillary teeth small, equal, 17 to 31 on each maxilla; head not distinct from neck; body cylindrical, tail long; scales smooth, in 15 to 17 longitudinal rows at mid-body; apical pits absent; ventrals rounded; subcaudals paired; anal plate single or divided; eye large; hemipenes unforked, large spines present in proximal portion of organ; hypapophyses not present on posterior dorsal vertebræ.

Remarks.—The genus *Liopeltis* was established by Fitzinger (1843) to accommodate a single species, *Herpetodryas tricolor* Schlegel. At the same time he also proposed the nominal genus *Gongylosoma* for *Coronella baliodeira* Schlegel. Duméril (1853), and Duméril, Bibron, and Duméril (1854) established the nominal genus *Ablabes,* and included among other species *Coronella baliodeira* Schlegel. Boulenger (1890) selected *C. baliodeira* Schlegel as type species of *Ablabes,* making it a junior objective synonym of *Gongylosoma* Fitzinger.

In 1858, Günther proposed the nominal genus *Cyclophis.* To this genus he referred, among others, the nominal species *Herpetodryas tricolor* Schlegel and *Coluber aestivus* Linnæus. The latter was subsequently selected type species of the genus by Schmidt and Necker (1936). *Cyclophis,* thus, is a junior objective synonym of *Opheodrys* Fitzinger (see below). Günther, in 1862, also proposed the nominal genus *Phragmitophis* to which he assigned one species, *Cyclophis tricolor. Phragmitophis* is a junior objective synonym of *Liopeltis* Fitzinger.

In 1894a, Boulenger, who uniformly rejected all Fitzinger's names, included under the generic name *Ablabes* ten species of snakes, two from North America, and eight from China and southeastern Asia. Cope (1898), Taylor (1922), and Werner (1929) accepted Boulenger's arrangement of the genus though Cope and Taylor used Fitzinger's earlier name *Liopeltis.*

In 1935, Pope reexamined the generic affinities of the Chinese (including Formosa and the Ryukyu Islands), North American, and Oriental species of *Liopeltis.* He concluded that the Chinese and North American species were congeneric and were generically distinct from the Oriental forms. He referred the Chinese and North American species to the nominal genus *Eurypholis* Hallowell, the type species of which is *Eurypholis semicarinatus* Hallowell from the Ryukyu Islands. The Oriental species, including *H. tricolor* and *C. baliodeira,* were retained in *Liopeltis.* Schmidt and Necker (1936) pointed out that Pope was in error in using *Eurypholis* Hallowell because this

name was preoccupied by *Eurypholis* Pietet proposed for a group of fossil fish in 1852. They substituted *Opheodrys* Fitzinger to accommodate the North American and Chinese species which Pope believed were congeneric.

Bourret (1936) reverted to the Boulengerian arrangement of *Liopeltis*, including under this name Chinese, North American, and Oriental species. However, Smith (1943) accepted the arrangement proposed by Pope, as did Haas (1950).

As understood by Smith (1943) and Haas (1950) the genus *Liopeltis* included among other species both *Herpetodryas tricolor* and *Coronella baliodeira*. In the course of this study seven of the ten species presently attributed to *Liopeltis* were seen, including those two. It is clear that the genus as presently constituted (Smith, 1943) is a heterogenous assemblage of species. Available data suggest the species may be divided into two groups, one typified by *C. baliodeira*, the other by *H. tricolor:*

C. baliodeira	*H. tricolor*
Scales in 13 rows at mid-body.	Scales in 15 to 17 rows.
Diameter of eye/head length less than 5.4.	Diameter of eye/head length more than 5.4.
Snout length/diameter of eye less than 1.4.	Snout length/diameter of eye more than 1.6.
Spines in proximal portion of hemipenes not enlarged.	Spines in proximal portion of hemipenes enlarged.
Nasal shield divided.	Nasal shield single, rarely divided.

In addition to the above, there is a marked difference in the shape of the head between the two species. In *C. baliodeira* the head is short, deep, and convex in profile and the eye is very large. In *H. tricolor* the head is long, shallow, and flattened in profile. *Ablabes longicaudus* Peters, and *Ablabes scriptus* Theobald agree with *C. baliodeira*. *Ablabes philippina* Boettger, *Cyclophis calamaria* Günther, *Ablabes rappi* Günther, and *Cyclophis frenatus* Günther agree with *H. tricolor*. No specimens of *Ablabes stoliczkæ* Sclater or *Ablabes nicobariensis* Stoliczka have been seen.

The marked differences between the two groups suggest they are not congeneric. *Coronella baliodeira* and its relatives *A. scriptus* and *A. longicaudus* are hereby transferred to the genus *Gongylosoma* Fitzinger (type species *C. baliodeira*, by monotypy). *Liopeltis* is restricted to include *H. tricolor* and those

species most similar to it (see above). *Ablabes nicobariensis* and *A. stoliczkae* are temporarily retained in *Liopeltis* until their zoological status can be determined.

Two species of *Liopeltis* are recorded from the Philippines; both are found in the Palawan Archipelago. *Liopeltis tricolor*, known from Palawan Island in the Philippines, is widely distributed throughout western Indonesia; *L. philippina* is known only from the Philippines where it is found on the Calamian Islands and on Palawan. This latter species is obviously derived from *L. tricolor*. The two forms may be separated by at least one distinctive morphological character and by color pattern. Although similar morphologically, the two species appear to be reproductively isolated as evidenced by the occurrence of both forms in the Puerto Princesa region of Palawan Island without the appearance of intergrades.

Key to the Philippine species of Liopeltis

1*a*. Nasal shield fused to internasal; four brown longitudinal stripes on dorsum ..... *L. philippina*

1*b*. Nasal shield not fused to internasal; dorsum uniform above, without longitudinal stripes ..... *L. tricolor*

LIOPELTIS PHILIPPINA (Boettger).

Ablabes philippinus BOETTGER (1897) 164 (Culion and Samar islands; type locality designated as Culion Island [see below]; type in Senckenberg Museum; original description), (1898) 78 (listed); GRIFFIN (1911) 261 (Palawan Is. [Iwahig], listed in key); WERNER (1929) 152, 153 (listed).

Liopeltis philippinus Taylor (1922) 164, pl. 20 (synonymy; description; distribution compiled).

Range.—BUSUANGAO. CULION. PALAWAN: Puerto Princesa [*]; Iwahig.

Material examine (2).—BUSUANGA: (CAS 15284). PALAWAN: Puerto Princesa (CNHM 53372).

Taxonomic notes.—This species is similar to *L. tricolor*, differing in relatively minor, but apparently constant characters. Both species occur on northern Palawan Island. Otherwise, *L. philippina* is known from the Calamianes group of islands north of Palawan. Boettger records one of the syntypes as coming from Samar Island. However, the record is seriously doubted, and it seems likely the locality datum accompanying the specimen is in error. Inasmuch as the locality datum accompanying the

[1] Boettger's [(1897) 164] Samar record for this species is doubted.

second of the two paratypes, *i.e.* Culion Island, agrees with other data available on the distribution of this animal, I hereby designate Culion Island, Calamianes group, type locality of *L. philippina*.

TABLE 1.—*Summary of variation between sexes in Liopeltis philippina.*

Character	Male	Female
Ventrals	139–140	150
Subcaudals	111 119	110
Tail length/standard length	0.64	0.55

Diagnosis.—Scales in 15 longitudinal rows at mid-body; loreal absent; nasal shield single, anterior portion united with internasal; dorsum with four dark brown longitudinal stripes; subcaudals 110 to 119. Measurements: Body (♂) 341 mm, (♀) 335 mm; tail (♂) 219 mm, (♀) 183 mm.

Descriptive notes.—Maxillary teeth 29; posterior portion of nasal divided by horizontal suture which arises from center of nostril and extends to posterolateral border; 8 upper labials, fourth and fifth border orbit; 1 preocular; 2 postoculars; temporals 1 + 2; body dorsal scales reduce: 15 (3 + 4 [84–92]) 13; ventrals, 139 to 150; subcaudals, 110 to 119; anal plate divided.

Hemipenes extend to seventh subcaudal plate, unforked; sulcus spermaticus unforked; proximal half with well-developed spines which become somewhat larger toward middle of organ; spines abruptly become very small at middle of organ and cover entire distal half; no flaplike ridge in distal portion as in *L. tricolor*.

Color (in alcohol) light grayish tan to olive above; two dark olive brown dorsolateral stripes originate just behind head and extend onto tail, and two lateral olive brown stripes which originate just behind the eye extend to near anus.

Sexual dimorphism.—Data from three specimens, two of which were examined here and one recorded by Taylor [(1922) 164], suggest the sexes may differ in ventral counts and tail length/standard length ratio (Table 1). The position of reduction of the body scales from 15 to 13 may also be subject to sexual dimorphism. Data from two specimens are as follows: (♂) 15 (3 + 4 [91–92]) 13; (♀) 15 (3 + 4 [84–85] 13.

Inter-island variation.—Neither is there evidence of geographic variation in the sample available for study nor is it indicated by material recorded in the literature.

LIOPELTIS TRICOLOR (Schlegel).

Herpetodryas tricolor SCHLEGEL (1837) 187, pl. 6, figs. 16–18 (type loc: Java; type in Leiden Museum; original description).

Liopeltis tricolor COPE (1860) 559 (listed as type species of genus); JAN (1869) Livr. 31, pl. 6, fig. 2; TAYLOR (1922) 162, pl. 11, figs. 3–5, pl. 19 (Bubuan Is; Palawan Is. [Iwahig, Taytay]; synonymy, description, variation, material examined, measurements and counts); Haas (1950) 560 (distribution compiled).

Phragmitophis tricolor GÜNTHER (1862) 126 (explanation for transferring *H. tricolor* to the new genus).

Ablabes tricolor BOULENGER (1894a) 281 (distribution compiled; synonymy, description, counts of material examined); FLOWER (1899) 673 (Singapore); GRIFFIN (1909) 599 (Palawan Is. [Iwahig]; listed), (1911) 201 (Palawan Is. [Iwahig]; listed in key, distribution compiled); BOULENGER (1912) 151 (distribution compiled; description); DE ROOIJ (1917) 138 (distribution compiled; description); TAYLOR (1918) 260 (Bubuan Is. [Tapian Group]; counts and measurements); WERNER (1929) 152, 153 (distribution compiled; listed in key).

Gongylosoma tricolor SWORDER (1923) 64 (not seen).

Range.—(Philippine localities only.) BUBUAN. PALAWAN: Iwahig *; Taytay.

Material examined (7).—PALAWAN: Iwahig (CAS 62168: CNHM 15054); without exact locality (CM 8810). BORNEO: Sarawak: Limbang District (MCZ 11270). Without exact locality: CNHM 72453. JAVA: Buitenzorg (USNM 43477); Kosterm (CNHM 72453).

Taxonomic notes.—Specimens from Palawan do not differ from those from Java and Borneo. *Liopeltis tricolor* is treated as a monotypic species.

Diagnosis.—Scales in 15 longitudinal rows at mid-body; loreal absent; nasal shield single, not united with internasal; dorsum monochromatic; subcaudals 103 to 133. Measurements: Body (♂) 330 mm, (♀) 281 mm; tail (♂) 205 mm, (♀) 167 mm.

Descriptive notes.—Maxillary teeth 28 to 31; 8 or 9 upper labials, fourth and fifth or fifth and sixth border orbit; 1 preocular, rarely 2; 2 postoculars: temporals 1 + 2; body dorsal scales reduced; 15 (3 – 4 [91–114]) 13; caudodorsal scales reduced: 6 (2 + 3 [9–161]) 4; ventrals 140 to 187; subcaudals 103 to 133; anal plate single.

Hemipenes extend to from fifth to eighth subcaudal plate, unforked; sulcus spermaticus unforked; proximal half with large spines; distally there is a prominent flaplike ridge beset with small spines; one large spine, two or three caudal plates in length, arises at juncture of large and small spines.

Color (in alcohol) deep brown above which gradually becomes lighter posteriorly; a black stripe originates on nasal, extends through eye, and passes onto body along second scale row for about two head-lengths behind head; immaculate cream below.

Sexual dimorphism.—A single female specimen was available for study. Literature records for four additional specimens said to be females were available; the sexing data are accepted with reservation. Five male specimens were examined, and literature records of five specimens said to be males were available.

Combining the author's data with those abstracted from the literature, the following ranges for ventrals and subcaudals are obtained:

	Male	Female
Ventrals	140–160	148–187
Subcaudals	116–133	103–133

The tail length/standard length ratio does not appear to differ between the sexes. The ratio for the five males examined here ranged from 0.58 to 0.66 (mean–0.631); the ratio for the single female was 0.60. The hemipenes are short structures, occupying less than 7 per cent of the tail, so it is not too surprising there should be little difference in the tail length/standard length ratio between sexes.

Inter-island variation.—There are indications of inter-island variation in ventral and subcaudal counts. Specimens from Borneo differ from both Javanese and Palawan individuals in their greater number of ventral and subcaudal shields. On the other hand, Palawan specimens appear to have shorter tails than specimens from Borneo or Java. Data for the above-mentioned characters are summarized in Table 2.

Ecological notes.—Little is known about the habits of these snakes. They are rather elongate, slender snakes, and presumably are arboreal. One specimen, indeed, was collected in a low tree [Taylor (1918) 260, (1922) 163].

Insect remains were found in the stomachs of three specimens. Stoliczka [(1873) 122] reported a spider from the stomach of

a Sumatran individual. In one specimen examined here, a few ventral plates of some snake were found in the stomach.

TABLE 2.—*Summary of inter-island variation in several characters in Liopeltis tricolor.*

Island	Ventrals			Subcaudals		Tail length/ standard length
	N	Males	Females	Males	Females	
Palawan	4	149(3)	148(1)	116–125(3)	121(1)	0.55–0.61(4)
Borneo	6	153–160(4)	167–171(2)	124–133(4)	127 133(2)	0.60 0 62(2)
Java	4	140–149(3)	137(1)	118 131(3)	108(1)	0.65–0.66(2)

NOTE: Number in parenthesis indicates the number of specimens for which data were available.

Genus SIBYNOPHIS (Fitzinger)

Sibynophis FITZINGER (1843) 26 (type species *Herpetodryas geminatus* Schlegel, by monotype).

Enicognathus [nec Gray (1840) Aves] Duméril, Bibron, and Duméril (1854) 328 [type species *Herpetodryas geminatus* Schlegel, by subsequent selection by Stejneger (1910) 102].

Henicognathus [nec Agassiz (1846); emendation of *Enicognathus* Gray (1840) Aves] COPE (1868) 132 paragraph 1, line 1 *et seq* (erroneous subsequent spelling of *Enicognathus*).

Polyodontophis BOULENGER (1890) 301 (type species *Herpetodryas geminatus* Schlegel, by reason of its status as a substitute name for *Enicognathus*).

Definition.—Teeth numerous, 35 to 48 on each maxilla, subequal or the first and last 2 or 3 slightly smaller than the rest; maxillary bone extends a distance of 8 to 14 teeth beyond ectopterygoid-maxillary articulation; maxillary process of palatine broad, articulates with palatine process of maxilla; no dorsomedial extension of palatine present; pterygoid not broad, without strongly-developed dorsolateral ramus; dentary bone free from posterior part of articular; compound bone (angulare and articulare) curves outward at its anterior end and joins dentary bone at an acute angle; body cylindrical, tail long; scales smooth, without apical pits, in 17 longitudinal rows throughout (except in *S. subpunctatus* in which there is a reduction to 15 scale rows just in front of the anal plate); ventrals rounded; subcaudals paired; anal plate divided; pupil round; hemipenes unforked, spinose throughout, calyculate distally; hypapophyses present on posterior vertebræ.

Remarks.—Until recently the genus *Sibynophis* included a number of widely scattered species inhabiting Madagascar, southeastern Asia, and Central America. Taylor and Smith (1943) proposed the American species be separated into a distinct genus, *Scaphiodontophis*. In 1956, Leviton and Munsterman found reason to refer the Malagasian species to a new genus *Parasibynophis* which Guibé [(1958) 205] suggested was a synonym of the Malagasian genus *Liophidium*. The genus *Sibynophis* is restricted to include a group of southeast Asian snakes only. Seventeen nominal species are now referred to it. Nine are currently recognized; one, *Sibynophis bivittatus*, enters the Philippines.

The structure of the lower jaw among the sibynophine snakes deserves note. A number of colubrid genera have been shown to possess modified lower jaws. In some, for example *Dipsas*, *Sibon*, and *Trachymenis*, the dentary bone is loosely articulated posteriorly with the compound bone (articulare and angulare). In *Sibynophis*, *Scaphiodontophis* and *Parasibynophis* [= *Liophidium*, fide *Guibé*, 1958], the posterior portion of the dentary bone is completely free from the compound bone. In fact, the compound bone joins the dentary at a point about one-third the distance from the anterior end of that bone and at an acute angle to its longitudinal axis. In *Dipsas*, the dentary lies immediately above the compound and the two are aligned along the same longitudinal axis. The situation in *Dipsas*, thus, does not differ strikingly from the normal colubrid condition.

Dipsas and *Sibynophis* agree with each other, and therefore differ from most other colubrid snakes, in two respects: (*1*) a slip of muscle, derived from the *adductor mandibulare* (= *adductor externis superficialis* la [Haas (1931)] and *adductor dentalis* [Dunn (1951)] inserts on the posterior end of the dentary (this condition, which is not common among colubrid snakes, presumably affords additional control for vertical movement of the dentary), and (*2*) the pterygoid bone extends beyond or does not attain the quadrate and is not attached to the quadrate by any ligamentous connection, which is the usual condition in colubrid snakes. Dunn [(1951) 355] suggested that this modification might permit independent movement of each of the four elements comprising the upper and lower jaws [see also Peters (1960) 15–17].

Dunn theorized that in *Sibon* and *Dipsas* the modifications of the lower jaw indicate changes primarily concerned with food

habits. Schmidt (1950) offered a similar explanation for *Sibynophis*. *Dipsas* and *Sibon* are known to feed upon soft-bodied organisms, especially snails and slugs, and the modifications noted may have some special significance in aiding the snake to engulf these foods. *Sibynophis* and its relatives subsist largely upon skinks. It may be that independent movements of the toothed bones permit the snake to grasp these smooth, rounded and "tough-skinned" animals in a viselike manner at any one moment with three of its four dentaries while the fourth is moved forward to a new position.

In the structure of their teeth, the sibynophine snakes differ from other colubrid groups. In *Sibynophis* and its relatives, the teeth are compressed, the anterior-medial edge being knife-like, the tip is somewhat rounded and the whole tooth only slightly recurved. The maxillary and mandibular teeth, among other colubrid genera, are strongly recurved, are generally cone-shaped, and terminate in needlelike points.

There seems to be other peculiarities in the dentition of the sibynophine snakes such as mode of replacement, which does not seem to follow the alternate pattern common to the colubrid snakes, but these have not been studied.

SIBYNOPHIS BIVITTATUS (Boulenger).

Polyodontophis bivittatus BOULENGER (1894b) 82 (type loc: Palawan Island; type in British Museum; original description), (1896) 597 (description quoted); BOETTGER (1898) 17 (Culion Island; listed); GRIFFIN (1909) 596 (listed), (1911) 256 (listed in key); THOMPSON (1913) 423 (describes hemipenes; suggests relationship to Colubrinæ rather than Natricinæ).

Sibynophis bivittatus TAYLOR (1922) 80, pl. 10, fig. 1 (Palawan Island, Busuanga Island [Minuit]; synonymy, description, variation, counts and measurements of material examined; WERNER (1929) 7 (listed in key); LEVITON and MUNSTERMAN (1956) 2 (listed).

Range.—BUSUANGA: Minuit. CULION: without exact locality *. PALAWAN: Brooke's Point *.

Material examined (6).—BUSUANGA: without exact locality (CAS 60548). CULION: without exact locality (CNHM 53368). PALAWAN: Brooke's Point (MCZ 25609); without exact locality (CNHM 53367, 53369). PHILIPPINES: without exact locality (CAS 15205).

Taxonomic notes.—This species appears to be endemic to the Palawan Archipelago. It is similar to *S. geminatus* of Indonesia. Unfortunately no specimens of *S. geminatus* from

Borneo have been available; the sample of *S. bivittatus* has been compared with specimens of *S. geminatus* from Java. The following major differences are evident: (*1*) a lower number of upper labials in *S. bivittatus* (usually eight rather than nine); (*2*) upper labials three to five (rarely four to five) border the orbit in *S. bivittatus*, four to six in *S. geminatus;* and (*3*) a white interocular bar is present in *S. bivittatus*, a white nuchal bar is present in *S. geminatus*. *Sibynophis g. insularis* Mertens agrees with *S. bivittatus* in the number of upper labials and the upper labials which border the orbit; they differ in coloration and dentition. I suspect that material from Borneo will be intermediate in character between *S. bivittatus* and Javanese *S. geminatus*.

Taylor's statement [(1922) 80], "The occurrence in the Philippines of a species of this genus [*Sibynophis*] is somewhat unusual, as no other member of the genus appears to have been discovered in any of the East Indian Islands," is without foundation. F. Boie (1827) described *S. geminatus* from Java, and the species has been recorded from Sumatra and Borneo by several authors [Boulenger (1893) 185; de Rooij (1917) 54].

Diagnosis.—Eight upper labials (rarely nine), the third to fifth or fourth and fifth bordering the orbit; two anterior temporals; white interocular bar present; no light nuchal bar; two narrow, white dorsolateral stripes present on dark background; subcaudals (?93) 110 to 112.

Descriptive notes.—Maxillary teeth 39 to 43; nasal shield small, divided, nostril small; loreal slightly longer than high; 1, rarely 2, preoculars; 2 postoculars, one or both of which are in contact with parietal; 8 upper labials, rarely 9, the third, fourth, and fifth, fourth and fifth, or fourth, fifth, and sixth bordering the orbit; temporals 2 + 2, the lower anterior shield in contact with the seventh and eighth upper labials: 9 lower labials, the first pair in contact behind the mental; caudodorsal scales reduced: 6 (2+ 3 [16–21] 4 (1 + 2 [86–96]) 2; ventrals 145 to 155; subcaudals (?93) 110 to 112; anal plate divided.

Hemipenes extend to the eleventh subcaudal plate, unforked; sulcus spermaticus unforked; basal portion with convoluted ridges beset with minute spines; proximal half spinose, with one very large spine flanked by two slightly shorter spines and followed by one or two transverse rows of moderate spines; distal half spinose, with very small irregularly-shaped calyces

having scalloped edges bearing minute spines at distal end; a prominent spongy, spinose ridge in distal half is coincident with the large spine in the proximal portion.

Color (in alcohol) dark brown above, which extends onto outer edge of ventrals or just to outer row of dorsal scales; a distinct white (while in life, too) dorsolateral stripe, occupying parts of the fourth, fifth, and occasionally sixth scale rows, originates either behind the eye or the parietals, and extends the length of the body and the tail; a distinct broad white [light salmon red in life [*fide* Taylor (1922) 82] interocular bar present; upper labials white, bordered above by a black stripe that passes through the eye; temporal region and internasals lack dense melanin pigment and are lighter than ground color. Venter immaculate creamy white (greenish yellow in life).

Ecological notes.—A single skink (unidentified) was found in the stomach of one individual. Skinks appear to form the principal, if not the only food taken by members of the genus [Wall (1921) 85; Pope (1935) 84, 87, 432; Smith (1943) 278; Taylor and Smith (1943) 304, 327; Leviton and Munsterman (1956) 7].

Nothing is known of the other habits or habitat of this species. Most species of *Sibynophis* appear to inhabit hilly regions at moderate or low altitudes. At least two species, *S. collaris* and *S. chinensis grahami*, are found at altitudes in excess of 5,000 feet (10,000 feet in the case of *S. collaris* from Nepal). One of the specimens of *S. bivittatus* collected by Taylor was taken at the seashore on Busuanga Island. Data for the other specimens are not available.

All species whose breeding habits are known are oviparous, laying two to five eggs (*S. collaris, S. chinensis,* and *S. subpunctatus*).

Remarks.—All specimens seen here were males. None of the specimens of *S. bivittatus* reported in the literature were sexed. Therefore, measurements and counts for females are not available for comparison.

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NEW OR LITTLE-KNOWN TIPULIDÆ FROM EASTERN ASIA (DIPTERA), LII

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THREE PLATES

At this time I am continuing the consideration of the crane flies taken on the Third Archbold Expedition to New Guinea, covering especially the Snow Mountains in the former Netherlands New Guinea, now called West Irian or West Papua. A short account of this expedition, with a list of the seventeen collecting stations established by the entomologist, Dr. Lambertus Johannes Toxopeus, has been provided in another paper which may be consulted. [Alexander (1958) 657–658.] Other papers that consider the expedition and bear upon the Tipulidæ that were secured are provided in the references. (See references on page 417.)

As has been indicated in these earlier papers, the main series of specimens resulting from this expedition and taken by Dr. Toxopeus is preserved in the Government Museum of Natural History, Leiden, Netherlands, with duplicates where available being deposited in the collection of the American Museum of Natural History, New York, and in the author's collection. I am most deeply indebted to Director M. A. Lieftinck, of the Leiden Museum, for the privilege of retaining the types of uniques in my personal collection.

LIMONIINÆ
LIMONIINI

LIMONIA (LAOSA) PAVO sp. nov. Plate 1, fig. 1.

Size small (wing of female 9 millimeters); general coloration of thorax obscure brownish yellow, darker behind; antennal flagellum orange; halteres pale yellow, apex of knob dark brown; wings subhyaline, strikingly patterned with pale brown and brownish black, the basal areas ocelliform; a broad complete crossband beyond cord; supernumerary crossveins in cells R_1 and R_5.

Female.—Length, about 7.5 millimeters; wing, 9.

Rostrum black; palpi dark brown. Antennæ with scape pale brown, pedicel and flagellum dull orange, the outer segments slightly darker; basal flagellar segments oval, with short verticils. Head light gray, posterior vertex with a narow central brownish black line that narrows to a point in front; anterior vertex reduced to a narrow strip that is about equal in width to two rows of ommatidia.

Pronotum and cervical region yellowed, weakly darker above. Mesonotal præscutum obscure yellow, darker on sides behind the humeri; posterior sclerites of notum yellowish brown, the sides of scutal lobes darker. Propleura yellow, remainder of pleura darker brownish yellow. Halteres yellow, apical half of knob dark brown. Legs with coxæ and trochanters yellow, apex of latter narrowly blackened and with a microscopic spinous point; remainder of legs broken. Wings (Plate 1, fig. 1) with the ground subhyaline, strikingly patterned with very pale brown and brownish black; the paler areas include a complete band beyond cord, broadest in front between tips of veins Sc and R_{1+2}, narrowed behind at end of vein 1st A, narrowest over outer end of cell 1st M_2, the band narrowly bordered by darker brown; other paler markings include ocelliform areas before cord, the most complete at origin of Rs with a more basal one semicircular in outline, both of these narrowly dark brown on outer part, bordered internally by a subequal white ring; still paler areas in outer radial field over the supernumerary crossveins, connected along vein R_{4+5}, with the central crossband and near end of vein 2nd A, with other marks at ends of veins R_3, R_{+5}, M_{1+2}, M_3 and M_4, extended basad and expanded into the cells to form an oblique subterminal band; the darkest areas are restricted brownish black marks on anterior cord with a further large circular spot in cell R adjoining Rs; veins yellow, darker at the anterior cord. Venation: Sc_1 ending some distance beyond level of fork of Rs, Sc_2 near its tip; supernumerary crossveins in cells R_2 and R_5; m-cu at near midlength of M_{3+4}; anal veins bent very strongly into the margin; cell 2nd A broad.

Abdominal tergites bicolored, dark brown, with pale yellow bases, outer segments uniformly darkened; basal sternites obscure yellow, the outer segments darker. Ovipositor with cerci slender, the tips acute.

Habitat.—West Papua.

Holotype, female, Sigi Camp, altitude 1,350 meters, February 20, 1939 (*Toxopeus*); Alexander Collection, through Dr. Lieftinck.

Limonia (*Laosa*) *pavo* is readily told from all other regional members of the subgenus having two supernumerary crossveins in the wing by the striking and distinctive wing pattern. The most similar such species include *L.* (*L.*) *falcata* Alexander and *L.* (*L.*) *innuba* Alexander, readily distinguished by the coloration of the wings.

LIMONIA (LAOSA) SUFFALCATA, sp. nov. Plate 1, fig. 2.

Size large (wing of male 11 millimeters); mesonotum ashy gray, patterned with brown; femora light yellow, tips dark brown, tibiæ yellow, bases very narrowly darkened; wings barely subfalcate, subhyaline, patterned with brown, the major crossbands pale brown, narrowly bordered by darker brown; cell 2nd A broad.

Male.—Length, about 8 millimeters; wing, 11.

Rostrum brownish black; palpi black. Antennæ with scape brownish black, pedicel yellow; flagellum broken. Head dark brown, anterior vertex more pruinose, posteriorly reduced to a capillary strip or with eyes virtually contiguous.

Pronotum darkened. Mesonotal præscutum with the ground clear ashy gray, greatly restricted by a brown central stripe that extends laterad to the margin, limiting the ground to the humeral triangles and a broad area before suture; scutum chiefly dark brown, including the midregion; scutellum ashy gray, the center of disk medium brown; postnotum dark brown, the mediotergite with a pair or oval discal light gray spots. Pleura of type crushed, apparently variegated dark brown and light gray, the latter color chiefly on the ventral sternopleurite and posteriorly; dorsopleural membrane pale. Halteres light yellow, knob dark brown. Legs with coxæ medium brown, the middle pair more pruinose; trochanters obscure yellow; femora light yellow, tips dark brown; tibiæ yellow, bases very narrowly darkened; tarsi yellow, the three outer segments brownish black; claws with a major outer tooth and a more basal comblike row of about seven smaller teeth. Wings (Plate 1, fig. 2) subhyaline, patterned with brown, including a narrow band at near one-fourth the length, and a larger irregular area beyond midlength, both of these pale brown variegated with paler and narrowly margined with darker brown; basal band narrowed posteriorly,

ending at vein 2nd A; the complex outer band appears as a narrow seam over the cord, more or less confluent with a much broader one beyond, both bands connected in cell M_4; in outer radial field with still other darkened areas, confluent with one another and with the major band in cell R_5; small paler marks at ends of veins M_{1+2}, M_3, M_4 and 1st A; in the region of the arculus with a restricted yellowed mark that is narrowly bordered by dark brown; veins yellow, the color persisting in the darkened areas and appearing conspicuous against this background. Wing tip weakly subfalcate. Venation: R_2 lying basad of free tip of Sc_2; supernumerary crossveins in cells R_4 and R_5, the latter more basad; cell 1st M_2 relatively short, strongly widened outwardly, m-cu at near midlength of M_{3+4}; vein 2nd A strongly arched, the cell broad.

Abdomen short, dark brown, the extreme posterior borders of the intermediate tergites yellow, broader on the intermediate sternites; hypopygium dark brown.

Habitat.—West Papua.

Holotype, male, Rattan Camp, altitude 1, 150 meters, February 13, 1939 (*Toxopeus*); Alexander Collection.

Limonia (*Laosa*) *suffalcata* is most similar to *L.* (*L.*) *falcata* Alexander, of New Britain and Northeast New Guinea, differing in the feebly falcate wings, details of venation, and lack of brown dots in the wing cells.

LIMONIA (LIBNOTES) INVICTA sp. nov. Plate 1, fig. 3; Plate 2, fig. 21.

Size relatively large (wing 13 millimeters or more); mesonotal præscutum chestnut brown, scutal lobes dark brown; wings faintly tinged with brown, conspicuously patterned with darker brown; Sc very long, Sc_1 ending beyond level of r-m; m-cu at or beyond midlength of M_{3+4}, male hypopygium with tergite broadly emarginate; ædeagus at apex produced into acute nearly hyaline points.

Male.—Length, about 9 to 10 millimeters; wing, 13 to 14.

Female.—Length, about 10 to 11 millimeters; wing, 13.5 to 14.

Rostum brownish black, mouthparts and palpi black. Antennæ with scape dark brown, remainder black; proximal flagellar segments short-oval, outer segments more elongate, terminal segment almost one-half longer than the penultimate. Head dark brown; anterior vertex narrow, less than the diameter of the scape.

Cervical sclerites and pronotum dark brown. Mesonotal præscutum chestnut brown to fulvous, the extreme cephalic border slightly darker; scutal lobes dark brown, central area and the scutellum testaceous yellow; postnotum obscure yellow. Anterior thoracic pleura dark brown, pleurotergite and posterior sclerites more yellowed. Halteres with stem yellow, apex of knob dark brown. Legs with coxæ and trochanters light yellow; femora dark brown, bases slightly darker; tibiæ and tarsi dark brown; claws of moderate length, with one major outer spine and three smaller more basal ones. Wings (Plate 1, fig. 3) faintly tinged with brown, patterned with darker brown, including the stigma and broad seams at origin of Rs, cord and outer end of cell 1st M_2; vague still paler clouds over outer veins, Cu and 2nd A; veins brown. Longitudinal veins beyond cord with macrotrichia, including also a few on Rs and distal ends of M, Cu_1 and the anals. Venation: Sc very long, Sc_1 ending slightly beyond the level of r-m, Sc_2 near its tip, faintly indicated; basal section of R_{4+5} short, in longitudinal alignment with Rs; cell 1st M_2 long, exceeding vein M_4; m-cu at or shortly beyond midlength of M_{3+4}.

Abdomen dark cinnamon brown, the outer segments, including hypopygium, brownish black. Ovipositor with cerci very slender, gently upcurved to the acute tips. Male hypopygium (Plate 2, fig. 21) with posterior border of tergite, *t*, broadly emarginate, the obtuse lobes with abundant long setæ. Ninth sternite narrowed outwardly, provided with long coarse setæ, those at base marginal, the outer ones slightly submarginal. Basistyle, *b*, with ventromesal lobe low and rounded. Dorsal dististyle a curved sickle, narrowed gradually to the acute tip; ventral style, *d*, with its area about one-half greater than that of the basistyle, setæ relatively short; rostral prolongation a compressed cleaverlike blade, on its face near base with a low blackened tubercle that bears a single rostral spine, with a much smaller normal bristle on its base. Gonapophysis, *g*, with mesal-apical lobe slender, the darkened tip slightly outcurved, lower margin before apex with a few low denticles. Ædeagus with each apical lobe produced into a nearly hyaline acute point (one-half shown in figure).

Habitat.—West Papua.

Holotype, male, Letterbox Camp, east of Mount Wilhelmina, 3,560 meters, September 12, 1938 (*Toxopeus*). Allotopotype, female, pinned with the type. Paratopotypes, males and fe-

males; paratypes, male and females, Scree Valley Camp, at foot of Mount Wilhelmina, 3,800 meters, September 14, 1938 (*Toxopeus*).

By Edwards's key to the Old World species of *Libnotes* (1928), *Limonia* (*Libnotes*) *invicta* runs to couplet 53, now known to include a large number of Oriental-Australasian species. It is well distinguished from these by its large size, venation, wing pattern, and details of hypopygial structure.

LIMONIA (LIBNOTES) TIBIOCINCTA sp. nov. Plate 1, fig. 4.

Allied to *amatrix;* size large (wing of male over 11 millimeters); mesonotal præscutum silvery gray with two light brown intermediate stripes; pleura white, with a narrow dark brown dorsal stripe; halteres with stem light yellow, knob dark brown; legs black, bases of fore femora obscure yellow, all femora with a narrow white ring beyond midlength; tibiæ with a similar annulus a short distance beyond the base; wings whitened, a conspicuous brown pattern; abdomen yellow.

Male.—Length, about 9.5 millimeters; wing, 11.5; antenna, about 2.

Rostrum brownish black, relatively long, nearly equal to remainder of head; mouthparts and palpi black. Antennæ with scape and pedicel brownish black, basal four or five flagellar segments subglobular, orange, outer segments oval, dark brown; terminal segment about one-half longer than the penultimate; verticils small and weak. Anterior vertex buffy, narrow, about one-third the diameter of scape; posterior vertex light gray, with long scattered setæ from conspicuous brown punctures.

Cervical region and propleura brownish black; pronotum greenish yellow. Mesonotal præscutum light silvery gray, clearest laterally; a pair of light brown intermediate stripes, narrowly confluent in front, slightly divergent behind; scutal lobes dark brown, median region brownish yellow; scutellum dark brown, paler on central part; mediotergite darkened anteriorly, the posterior end broadly reddish yellow, pleurotergite reddish yellow, more or less pruinose. Pleura silvery white below, more yellowed dorsally, with a narrow but conspicuous dark brown stripe extending from the fore coxæ to the wing root, involving especially the dorsal anepisternum. Halteres with stem light yellow, knob dark brown. Legs with fore coxæ darkened, as described, remaining coxæ and trochanters yellowed; femora black, all with a narrow white ring just beyond midlength, bases of fore pair restrictedly obscure yellow; tibiæ

black with a narrow white ring at slightly more than its own length beyond the base, the extreme genua whitened; tarsi black. Wings (Plate 1, fig. 4) with the ground whitened, costal border narrowly darker, yellowed beyond origin of Rs and especially above vein R_2; a conspicuous brown pattern, arranged as follows: Narrow seams over Rs and base of R_{2+3}, R_2 and R_{2+2}, outer end of cell 1st M_2 and for a considerable distance along the veins beyond; a continuous seam along vein Cu in cell M; anterior cord narrowly darkened; other marginal areas at ends of veins M_{1+2}, M_4 and anals, most extensive on the last; still further linear marginal seams in outer radial field, all of M_4 and in both anal cells; veins very pale yellow, difficult to see in the ground fields, conspicuously dark brown in the patterned areas. Venation: Vein Sc long, Sc_1 ending about opposite midlength of cell 1st M_2; veins beyond cord long; cell 1st M_2 elongate, m about one-half longer than the transverse basal section of M_3; m-cu lying far distad, about its own length before fork of M_{3+4}; anal veins moderately curved into the margin.

Abdomen almost uniformly brownish yellow, with greenish tints, hypopygium darker brown.

Habitat.—West Papua.

Holotype, male, Moss Forest Camp, near Lake Habbema, 2,800 meters, October 31, 1938 (*Toxopeus*); Alexander Collection.

Among the now numerous members of the *amatrix* group, *Limonia* (*Libnotes*) *tibiocincta* is most closely allied to *L.* (*L.*) *adicia* Alexander, differing most evidently in the pattern of the wings and legs, including the sub-basal white ring on the tibia.

LIMONIA (RHIPIDIA) DIONE sp. nov. Plate 1, fig. 5; Plate 2, fig. 22.

Size large (wing over 10 millimeters); antennæ black, subpectinate in both sexes; mesonotum cinnamon brown, grayish white laterally; pleura gray, with a narrow longitudinal dark brown stripe; halteres yellow; legs with femora yellow, with a broad brownish black subterminal ring, most evident on fore legs; wings whitish hyaline, with an abundant brown spotted and dotted pattern; Sc ending beyond origin of Rs; male hypopygium with six long rostral spines on ventral dististyle.

Male.—Length, about 8 to 8.5 millimeters; wing, 10.5 to 11; antenna, about 1.4 to 1.5.

Female.—Length, about 8.5 to 10.5 millimeters; wing, 11 to 13.

Rostrum and palpi black, the former about one-half the remainder of head. Antennæ black; basal flagellar segments subglobular, the ventral faces slightly more produced to appear subpectinate, apex abruptly narrowed into a short neck; outer segment more elongate, the terminal segment from about one and one-half to nearly twice the penultimate; the subpectinate condition is found in both sexes. Head gray; anterior vertex slightly narrower than the diameter of the scape.

Cervical region and pronotum brown to dark brown. Mesonotal præscutum rich cinnamon brown, laterally broadly grayish white, median region before suture slightly darker brown; scutal lobes dark brown, median region restrictedly whitened; posterior sclerites of notum brownish yellow. Pleura gray, with a narrow but conspicuous dark brown longitudinal stripe extending from the cervical region to the base of abdomen, passing beneath the root of the halteres. Halteres light yellow. Legs with coxæ small, dark brown, gray pruinose; trochanters brownish gray; femora yellow, with a broad brownish black subterminal ring, the narrow apex yellow, the darkened ring of fore legs broader and darker; tibiæ yellow, tips blackened, broader on fore pair; tarsi yellow, tips of proximal three segments darkened, outer segments uniformly blackened; claws with a single well-developed tooth. Wings (Plate 1, fig. 5) whitish hyaline, including base, conspicuously patterned with abundant brown spots in all cells, larger and slightly more intense along costal border; major dark areas at midlength of cell Sc, an irregular virtually common mark at origin of Rs and fork of Sc, fork of Rs, stigma, and marginal seams at Cu and the anals; spots in cells commonly circular and showing little tendency to fuse; veins yellow in the ground, darker in the more heavily patterned areas. Macrotrichia on longitudinal veins beyond general level of outer end of cell 1st M_2, with others on R_{2+3} and R_5, lacking on Sc, M, Cu and the anals. Venation: Sc relatively long, Sc_1 ending about opposite one-third the length of Rs; cell 1st M_2 subequal to vein M_{1+2} beyond it; m-cu at or close to fork of M.

Abdomen of male brown, darker laterally, outer segments more uniformly darkened, styli paler. In female, abdomen more yellowed, narrowly darkened along the sides. Male hypopygium (Plate 2, fig. 22) with tergite, *t*, narrowed outwardly, apex very shallowly emarginate; borders, especially the apex, broadly thickened. Basistyle, *b*, with ventromesal lobe obtuse,

setæ large. Dorsal dististyle only gently curved, widened beyond midlength, suddenly narrowed into a straight apical spine; ventral style, *d*, with its area about one-half greater than that of the basistyle; rostrum elongate, tip obtuse, with a concentrated group of six long yellow spines at near midlength of outer margin. Gonapophysis, *g*, with mesal apical lobe elongate, apex slightly curved, with a subterminal flange. Ædeagus broad, narrowed apically, terminating in two small lobes.

Habitat.—West Papua.

Holotype, male, Scree Valley Camp, at foot of Mount Wilhelmina, 3,800 meters, September 18, 1938 (*Toxopeus*). Allotype, female, Letterbox Camp, 3,600 meters, August 30, 1938. Paratopotypes, males and females, with the type and with the allotype; 1 male, Lake Habbema, near Mount Wilhelmina, 3,450 meters, August 16, 1938 (*Toxopeus*).

This attractive high-mountain species is quite distinct from the other regional members of the subgenus having subpectinate antennæ. The conspicuously spotted wing pattern and the six long rostral spines of the male hypopygium provide distinctive features.

LIMONIA (DICRANOMYIA) ENCHARIS sp. nov. Plate 1, fig. 6; Plate 2, fig. 23.

Belongs to the *tristis* (*liberta*) group; size large (wing 11 millimeters or more); general coloration of mesonotum light gray, the præscutum and scutum patterned with brown; knobs of halteres paling to obscure yellow; femora black; wings whitened, with a heavy and conspicuous brown pattern of spots and dots; Sc_1 ending beyond origin of Rs, cell 1st M_2 longer than any of the veins beyond it, m-cu before fork of M; male hypopygium with posterior border of tergite very strongly convex; basistyle with two setiferous lobes additional to the ventromesal one; rostrum of ventral dististyle short and stout, with two spines.

Male.—Length, about 7.5 to 8 millimeters; wing, 11 to 12.

Female.—Length, about 8 millimeters; wing 11.

Rostrum black, slightly elongated, about one-third the remainder of head; palpi black. Antennæ black, scape more pruinose; flagellar segments oval, well-separated; terminal segment slightly longer than the penultimate. Head brownish gray; anterior vertex subequal in diameter to the scape.

Pronotum light gray. Mesonotal præscutum light gray, with three pale brown stripes; scutum light gray, lobes pale brown; posterior sclerites of notum light gray. Pleura dark colored, light gray pruinose. Halteres with stem buffy yellow, knob blackened basally, the apex broadly obscure yellow. Legs with coxæ and trochanters black, heavily gray pruinose; femora black, tibiæ a little paler, tips narrowly blackened; tarsi brownish black. Wings (Plate 1, fig. 6) whitened, with a heavy and conspicuous brown pattern, including major areas at origin of Rs, stigma, fork of Rs, near outer end of cell R_3, together with especially large marginal areas in cells Cu, 1st A and 2nd A, the last at midlength of cell; additional small circular brown dots in cells R and M, with one to few others in various cells beyond the cord; veins brown. Venation: Sc_1 ending some distance beyond origin of Rs, in cases to about one-fourth the length of the vein, Sc_2 slightly removed; cell 1st M_2 large, exceeding in length any of the veins beyond it; m-cu from about one-third to one-half its length before fork of M; cell 2nd A broad, the vein sinuous.

Abdomen dark brown, in cases the tergites more chestnut brown, patterned laterally and on posterior border with darker, outer segments more uniformly darkened. Ovipositor with cerci very slender. Male hypopygium (Plate 2, fig. 23) with posterior border of tergite, *t*, very strongly convex, the extreme border at midlength with a group of three or four long setæ, with other stronger punctures scattered over the disk. Basistyle, *b*, relatively small, its area less than that of the ventral dististyle; ventromesal lobe conspicuous, narrowed to the blunt tip which bears numerous strong setæ; a long fingerlike lobe on face of style, tipped with few very long setæ; mesal apical angle of style produced into a smaller lobe bearing much shorter setæ. Dorsal dististyle gently curved and widened on outer half, very gradually narrowed into a long straight spine; ventral style, *d*, relatively large, the prolongation stout, its apex very obtuse, with numerous small setæ at and near tip; two relatively short rostral spines. Gonapophysis, *b*, very broad basally, its mesal apical lobe a short hook.

Habitat.—West Papua.

Holotype, male, Scree Valley Camp, at foot of Mount Wilhelmina, 3,800 meters, October 20, 1938 (*Toxopeus*). Allotopotype, female, September 18, 1938. Paratopotypes, males, September 22, 1938.

This conspicuous fly is entirely distinct from the other regional members of the *tristis* group in the abundantly spotted wing pattern.

LIMONIA (DICRANOMYIA) NIVEIFUSCA sp. nov. Plate 1, fig. 7; Plate 2, fig 24.

Belongs to the *punctulata* group; mesonotal præscutum with three reddish brown stripes, scutellum yellow, pleurotergite with a brown spot; antennæ short, black; knobs of halteres yellow; legs brown, femora with a darker terminal or nearly terminal ring; wings dusky, conspicuously patterned with darker brown and snowy white, appearing as alternate areas along the veins; cell 1st M_2 long, m-cu some distance before fork of M; male hypopygium with two very small rostral spines; mesal-apical lobe of gonapophysis simple, gently curved to the acute tip.

Male.—Length, about 7 to 8 millimeters; wing, 8 to 11.

Female.—Length, about 7.5 to 8 millimeters; wing, 10 to 11.

Rostrum brownish yellow to dark brown; palpi black. Antennæ black; flagellar segments short-oval to oval, exceeding their verticils in length. Head brownish gray; anterior vertex narrow.

Pronotal scutum brown, scutellum and pretergites yellowed. Mesonotal præscutum almost covered by three reddish brown stripes, the darker interspaces reduced to capillary lines, humeral region more yellow or grayish yellow; scutal lobes reddish brown, median region and the scutellum light yellow; mediotergite reddish brown, pleurotergite ventrally patterned with darker brown. Pleura reddish brown. Halteres with stem dusky, knobs conspicuously orange or yellow. Legs with coxæ brownish yellow; trochanters yellow; remainder of legs pale brown to brownish yellow, tips of femora and tibiæ and the outer tarsal segments darker; in cases, the femoral darkening is slightly subterminal and the tips of the tibiæ almost black; claws with a large subbasal tooth, with about four progressively smaller spines nearer base. Wing (Plate 1, fig. 7) with the ground dusky, conspicuously patterned with darker brown and snowy white, chiefly occurring as alternating areas along the veins and surrounding membrane to produce a striking effect; the darkened areas include the basal two-thirds of cells Sc and C, with major darker marks at origin of Rs and fork of Sc, on costa above the fork of Rs, and as darkenings at arculus and two areas on vein 2nd A; smaller but conspicuous similar marks at origin of Rs, interrupted at cord, outer end of cell 1st M_2, tips of R_{1+2} and

R_{2+3}, together with still smaller marginal seams at tips of remaining longitudinal veins; veins in the darkened areas dark brown, the intervening sections broadly snowy white. Veins beyond general level of outer end of cell 1st M_2 with macrotrichia, basad of this with trichia on R_1 and R_{2+3}, lacking on Sc, Rs, basal section of R_{4+5}, all veins comprising cell 1st M_2 and the basal veins. Venation; Sc moderately short, Sc_1 ending just beyond origin of Rs, Sc_2 before this origin; cell 1st M_2 very long, subequal to or slightly exceeding the distal section of M_{1+2}, m-cu its own length or more before fork of M; cell 2nd A broad, the vein bent strongly into the margin.

Abdomen chiefly yellowed, pleural membrane dark brown, outer sternites weakly darkened. Ovipositor with cerci slender, gently upcurved. Male hypopygium (Plate 2, fig. 24) with the tergite, *t*, transverse, posterior border very shallowly emarginate, the low lobes moderately thickned, each with about twenty delicate setæ. Basistyle, *b*, with the ventromesal lobe obtuse. Dorsal dististyle stout, the outer half slightly curved, tip suddenly narrowed into a long spine; ventral style, *d*, with its total area about one-half greater than that of the basistyle, provided with relatively sparse short setæ; rostral prolongation cleaver-shaped, with two unusually small slightly separated spines. Gonapophysis, *g*, with mesal-apical lobe simple, gently curved to the acute tip.

Habitat.—West Papua.

Holotype, male, Letterbox Camp, east of Mount Wilhelmina, 3,560 meters, September 12, 1938 (*Toxopeus*). Allotopotype, female, 3,600 meters, August 30, 1938. Paratopotypes, females; paratypes, 1 female, Lake Habbema, near Mount Wilhelmina, 3,225 meters, August 4, 1938; males and females, Scree Valley Camp, at foot of Mount Wilhelmina, 3,800 meters, September 15–22, 1938 (*Toxopeus*).

Limonia (*Dicranomyia*) *niveifusca* is quite distinct from the other described regional members of the *punctulata* group in the yellow knobs of the halteres, striking wing pattern, and details of venation. The hypopygial details, especially the rostral spines, likewise are distinctive.

LIMONIA (GERANOMYIA) ATYCHIA sp. nov. Plate 1, fig. 8; Plate 2, fig. 25

Size relatively small (wing of male 6 millimeters); rostrum of female about one-half the remainder of body, black; mesonotum fulvous, pleura more yellowed; legs light brown; wings

subhyaline, with a heavy brown costal pattern, the darkened areas more extensive than the interspaces; m-cu nearly its own length before fork of M; ventral dististyle of hypopygium with rostral spines long and slender, arising from a common basal tubercle.

Male.—Length, excluding rostrum, about 5 millimeters; wing, 6.

Female.—Length, excluding rostrum, about 6 millimeters; wing, 6.7; rostrum, about 2.9.

Rostrum black, in the holotype male with apex broken; in female relatively long, about one-half the remainder of body; palpi black. Antennæ black; outer flagellar segments elongate, verticils short. Head blackened, with a narrow gray central line extended backward to the occiput.

Pronotum fulvous brown. Mesonotum fulvous, darker posteriorly; pleura more fulvous yellow. Halteres dirty white, setæ of stem unusually long and conspicuous, especially those of ventral surface. Legs with coxæ fulvous; trochanters testaceous yellow; femora light brown, extreme tips slightly more yellowed; remainder of legs very light brown. Wings (Plate 1, fig. 8) with the ground subhyaline, with a heavy chiefly costal brown pattern, beyond the arculus including seven major areas, the ground interspaces in male wider than those of female; third dark area at origin of Rs, fourth over tip of Sc and fork of Rs, last area smallest, over R_{4+5}; other darkened areas over cord, outer end of cell 1st M_2 and apex of vein 2nd A, the last smaller in the type male; cells M, Cu and outer posterior cells with the ground weakly suffused; very small marginal clouds at ends of veins M_3, M_4, Cu_1 and 1st A; veins yellow, very slightly darker in the patterned areas. Macrotrichia on longitudinal veins beyond cord, including also the outer end of M and tip of 2nd A; costal fringe of male relatively long and conspicuous. Venation: Sc very long, Sc_1 ending about opposite fork of the long Rs, Sc_2 at its tip; basal section of R_{4+5} about two-thirds R_{2+3}; cell 1st M_2 subequal to distal section of M_{1+2}; m-cu nearly its own length before fork of M.

Abdominal tergites dark brown, sternites, especially the basal ones, more yellowed. Male hypopygium (Plate 2, fig. 25) with the tergite, *t*, transverse, posterior border with a U-shaped emargination, the lobes low and obtuse; discal setæ larger than those of the lobes. Basistyle, *b*, with ventromesal lobe relatively long, setæ small. Dorsal dististyle unusually long and slender, outer

half strongly curved to the long apical spine; ventral style, *d*, large, its area about twice that of the basistyle, setæ small; rostral prolongation relatively short, bearing two very long slender spines from a common basal tubercle that is subequal in size to the apex beyond it. Gonapophysis, *g*, with mesal-apical lobe slender, pale. Ædeagus, *a*, narrow, apical lobes obtuse.

Habitat.—West Papua.

Holotype, male, Sigi Camp, along Sigi River, 1,500 meters, February 26, 1939 (*Toxopeus*). Allotopotype, female, February 18, 1939.

The most similar regional species is the larger *Limonia (Geranomyia) rupex* Alexander, which differs further in the pattern of the wings and structure of male hypopygium.

LIMONIA (GERANOMYIA) DICRANOSTYLA sp. nov. Plate 1, fig. 9; Plate 2, fig. 26.

General coloration of mesonotal præscutum and scutum dark plumbeous gray, scutellum yellow, pleura reddish brown; legs brownish yellow, outer tarsal segments darker; wings subhyaline, stigma brown, vein Sc long, costal fringe of male very long and conspicuous; male hypopygium with rostral prolongation of ventral dististyle bearing a powerful arm that forks into two strong spines.

Male.—Length, excluding rostrum, about 6 millimeters; wing, 7.5; rostrum, about 3.

Rostrum relatively long, especially the palpi, dark colored. Antennæ with scape and pedical dark brown; flagellum broken. Head dark brown; anterior vertex about one-half wider than the diameter of scape.

Pronotum dark plumbeous. Mesonotal præscutum with three dark plumbeous gray stripes, the interspaces brown, humeri more yellowed; scutum dark plumbeous, including the midregion; scutellum abruptly yellow, parascutella pale brown; mediotergite blackened, pruinose, posterior and lateral borders more reddened, pleurotergite similarly reddened. Pleura reddish brown. Halteres pale, especially the stem. Legs with coxæ testaceous yellow, fore pair slightly darker; trochanters yellow; remainder of legs brownish yellow, outer tarsal segments darker. Wings (Plate 1, fig. 9) subhyaline, stigma oval, brown; very restricted darkenings at the supernumerary crossvein in cell Sc, origin of Rs and Sc_2; veins brown. Costal fringe of male very long and conspicuous, shorter at base and before apex, longest over the central half. Venation: Sc long, Sc_1 ending nearly

opposite midlength of Rs, Sc_2 shortly removed; m shorter than the gently arcuated basal section of M_3; m-cu close to fork of M.

Abdominal tergites and hypopygium brown, sternites paler. Male hypopygium (Plate 2, fig. 26) with the tergite, *t*, long, the posterior border conspicuously emarginate to form two rounded lobes that are densely provided with strong dark setæ. What seems to represent the ninth sternite is a strong central structure, expanded outwardly into lateral lobes that are provided with strong darkened setæ, the midarea membranous. Basistyle, *b*, about one-half as extensive as the ventral dististyle; ventromesal lobe simple. Dorsal dististyle small, strongly narrowed outwardly, apex decurved; ventral style, *d*, large and fleshy; rostral prolongation very large and complex, strongly sclerotized, outer half slender, gently curved; before base of outer margin with a powerful arm that divides outwardly into two spines, the inner one more strongly curved; at base of outer margin with a long rostral spine from a conspicuous basal tubercle. Gonapophysis, *g*, with mesal-apical lobe elongate, gently curved to the acute tip, the concave margin with microscopic hyaline points.

Habitat.—West Papua.

Holotype, male, Araucaria Camp, 800 meters, March 18, 1939 (*Toxopeus*); Alexander Collection.

Limonia (*Geranomyia*) *dicranostyla* is unique among the known regional species in the structure of the male hypopygium, particularly the rostral prolongation. Other species, including the most similar one, *L.* (*G.*) *tanytrichiata* Alexander, have the outer arm of the prolongation simple.

LIMONIA (THRYPTICOMYIA) ARCUS sp. nov. Plate 1, fig. 10.

General coloration of thorax above reddish brown, more yellowed behind and on the pleura; antennæ black, flagellum subnodulose, the segments oval, produced into conspicuous apical necks; proximal two-thirds of basitarsi black, the remainder of tarsi snowy white; wings subspatulate in outline, crystal hyaline, entirely unpatterned; distal section of vein R_1 merging with R_2 and thence curved into vein R_{2+3} without a trace of the spur of vein R_{1+2}.

Female.—Length, about 4.8 millimeters; wing, 5.5.

Rostrum brown, paler distally; palpi brownish black. Antennæ black throughout; flagellum subnodulose, the individual segments oval, with distinct apical necks; longest verticils uni-

laterally arranged, considerably longer than the segments. Head dark brown, gray pruinose.

Thorax almost uniformly reddish brown to reddish yellow, the mesonotal præscutum and scutum darkened, posterior sclerites of notum and the pleura more yellowed. Halteres elongate, brown, knobs brownish black. Legs with coxæ and trochanters testaceous yellow; femora brown, the bases restrictedly paler; tibiæ and about the proximal two-thirds of the basitarsi brownish black to black, the remaining tarsal segments snowy white. Wings (Plate 1, fig. 10) with the prearcular region slender, giving to the wing a subspatulate appearance; crystal hyaline, unpatterned, even the stigma lacking; veins black, delicate, very distinct against the ground. Venation: Sc short, Sc_1 ending a short distance before origin of Rs, Sc_2 slightly removed from the tip, Sc_1 alone subequal or a little longer than r-m; free tip of Sc_2 pale but distinct, beyond which point vein R bends in a curve into R_{2+3}, there being no sign of a spur of vein R_{1+2}, as found elsewhere in the subgenus; cell 1st M_2 long, subequal to or a little longer than the distal section of vein M_{1+2}; m-cu at near two-thirds the length of cell 1st M_2; no vestige of Cu_2; vein 2nd A relatively short, the cell narrow.

Abdominal tergites brownish black, sternites, especially the more basal ones, somewhat more brightened.

Habitat.—West Papua.

Holotype, female, Baliem River, at south end of the Grand Valley, 1,600 meters, December 13, 1938 (*Toxopeus*); Alexander Collection.

Limonia (*Thrypticomyia*) *arcus* is quite distinct from the other regional members of the subgenus, especially in the venation, the arched course of veins R_1 and R_2 being unique among such species. The most similar form is *L.* (*T.*) *microstigma* (Alexander) which differs in details of venation.

HELIUS (HELIUS) SUBANÆMICUS sp. nov. Plate 1, fig. 11; Plate 2, fig. 27.

Allied to *anæmicus;* size small (wing of male less than 5 millimeters); general coloration yellow; legs pale brown, outer tarsal segments more whitened; wings fulvous yellow, veins unusually glabrous; male hypopygium with apex of outer dististyle obtuse.

Male.—Length, including rostrum, 4.4 to 4.7 millimeters; wing, 4.1 to 4.8.

Female.—Length, including rostrum, 5.3 to 5.5 millimeters; wing, 4.8 to 5.

Rostrum obscure yellow, slightly more than one-half the remainder of head; palpi dark brown. Antennæ short; scape brown, remainder black; flagellar segments oval, subequal in length to their verticils. Head brownish yellow.

Thorax almost uniformly yellow. Halteres yellow. Legs with coxæ and trochanters yellow; femora and tibiæ pale brown, outer tarsal segments more whitened. Wings (Plate 1, fig. 11) fulvous yellow, prearcular and costal regions clearer yellow, stigma very pale brown; veins pale brown. Costal fringe of male long; veins unusually glabrous, beyond cord with about six or seven scattered trichia on distal section of R_{4+5}, two or three on last section of M_{1+2}; R_1 with an almost complete series of trichia but with only a few on R. Venation: Sc_1 ending shortly before fork of Rs; branches of Rs strongly divergent, cell R_2 at margin less than one-fifth R_3; cell 1st M_2 relatively small, a little shorter than vein M_4; m-cu shortly beyond fork of M.

Abdomen yellow, subterminal segment in male in cases more darkened to form a narrow ring; hypopygium yellow. Ovipositor with valves, especially the cerci, very long and slender. Male hypopygium (Plate 2, fig. 27) with the basistyle, *b*, elongate, simple. Dististyles, *d*, terminal; outer style slender, apex simple, obtusely rounded; inner style longer, the outer fourth slender, gently decurved; surface of style with setæ, those on lower margin elongate. Ædeagus short; gonapophysis prolonged into a straight beak, the tip acute, outer margin with pale membrane.

Habitat.—Papua; West Papua.

Holotype, male, Mist Camp, West Papua, Idenburg River, 1,800 meters, January 4, 1939 (*Toxopeus*). Allotopotype, female, pinned with the type. Paratopotypes, 1 male, 1 female, January 1–4, 1939. Paratypes, 1 male, Araucaria Camp, West Papua, 800 meters, March 18, 1939; 1 male, Rattan Camp, West Papua, 1,200 meters, February 9, 1939; 1 male, Sigi Camp, along Sigi River, West Papua, 1,500 meters, February 18,1939 (*Toxopeus*); 1 female, Goodenough Island, Papua, east slope, 900 meters, October 24–30, 1953 (*L. J. Brass*); Fourth Archbold Expedition, American Museum of Natural History.

The most similar species are *Helius* (*Helius*) *anæmicus* Alexander (Philippines, New Guinea) and *H.* (*H.*) *stolidus* Alexander (New Caledonia) which have the venation of the radial field generally as in the present fly, differing in the

venation and trichiation of the wings and in the structure of the male hypopygium.

HELIUS: RHYNCHOLIMONIA subgen. nov.

Characters strongly combining features of both the genera *Limonia* Meigen and *Helius* St. Fargeau. Front (Plate 2, fig. 28) produced into a short cylindrical rostrum with the mouthparts and palpi at extreme tip, as in *Helius*. Antennæ (Plate 2, fig. 28) 15-segmented; scape very short, less than the pedicel. Claws apparently weakly toothed at near midlength. Wings (Plate 1, fig. 12) with R_2 lacking; Rs forking far distad, the second section of Rs more than twice r-m, the outer radial branches nearly parallel to one another for most of their lengths. Other venational features more as in *Limonia* include the elongate subrectangular cell 1st M_2, atrophied vein Cu_2, and the course of the anal veins. Ovipositor elongate, generally as in *Helius*.

Type of subgenus.—*Helius* (*Rhyncholimonia*) *dicroneurus* sp. nov. (Australasian Region: Papuan Subregion).

The present group is based on a puzzling crane fly that strongly combines the generic characters of *Limonia* and *Helius*. The nature of the rostrum and the venation of the radial field are as in *Helius* but the venation of the remainder of the wing is just as definitely as that of *Limonia*. The discovery of the male sex presumably will confirm the assignment of the group with *Helius* rather than with *Limonia*.

HELIUS (RHYNCHOLIMONIA) DICRONEURUS sp. nov. Plate 1, fig. 12; Plate 2, fig. 28.

Size small (wing less than 5 millimeters); general coloration of thorax and abdomen yellow; wings whitened, with a conspicuous light and darker brown pattern.

Female.—Length, including rostrum, about 4.3 millimeters; wing, 4.6; rostrum, about 0.3.

Rostrum (Plate 2, fig. 28) black, palpi and mouthparts light yellow; structure as described under the subgenus; rostrum cylindrical, with short appressed setæ. Antennæ (Plate 2, fig. 28) 15-segmented; brownish yellow; flagellar segments oval, a little shorter than the verticils, terminal segment about one-half longer than the penultimate. Head pale brown; anterior vertex relatively narrow, subequal to the diameter of the scape.

Pronotum and anterior mesonotum yellowed, posterior sclerites slightly darker, surface of notum weakly pruinose; pleurotergite and pleura yellowed. Halteres relatively long, stem pale yellow, knob dark brown. Legs with coxæ reddened; tro-

chanters testaceous; femora yellow, tips rather narrowly but conspicuously dark brown; tibiæ and tarsi whitened. Wings (Plate 1, fig. 12) whitened, with a conspicuous pattern of light brown and brownish black streaks; pale pattern including a broken band at arculus, interrupted in cell 1st A; a major area at origin of Rs, with others at ends of vein R_{1+2} and outer end of cell 1st M_2, all of these markings vaguely bordered by slightly darker brown; a narrower brown seam at cord; conspicuous brownish black seams over outer radial forks and distal section of M_{1+2}, with less evident markings at ends of veins M_2 to 2nd A, inclusive; veins light brown, darker in the patterned parts. Abundant macrotrichia on virtually all veins, lacking on bases of M, Cu and Rs. Venation: Sc long, Sc_1 ending shortly before the level of r-m, Sc_2 near its tip; Rs very long, with r-m at near two-thirds the length, the second section being nearly three times r-m; branches of Rs subparallel except at outer ends; cell 1st M_2 elongate, exceeding any of the veins beyond it; anal veins bent strongly into the margin, the cells broad.

Abdominal segments weakly bicolored, chiefly yellow, the basal and lateral parts of the sclerites slightly darker, subterminal segments darkened; ovipositor and genital shield yellowed. Ovipositor with cerci and hypovalvæ elongate, as in typical *Helius*.

Habitat.—West Papua.

Holotype, female, Lower Mist Camp, 1,600 meters, January 15, 1939 (*Toxopeus*); Alexander Collection.

Helius (*Rhyncholimonia*) *dicroneurus* is entirely distinct from all other known members of the genus. In its general appearance it more resembles an unusually small species of *Limonia* belonging to subgenera such as *Libnotes* or *Laosa*.

HEXATOMINI

ELEPHANTOMYIA (ELEPHANTOMYODES) NIVEIPES sp. nov.

Size relatively large (wing of male 10.5 millimeters); mesonotal præscutum cinnamon brown, darker medially in front; legs black, tarsal segments two to four snowy white; wings with a brown tinge, costal border and narrow seams over cord darker brown; abdomen conspicuously bicolored, brownish black, the bases of the segments broadly yellow; hypopygium brownish yellow.

Male.—Length, excluding rostrum, about 8 millimeters; wing, 10.5; rostrum, about 5.

Rostrum black, slightly more than one-half the remainder of body. Antennæ black; flagellar segments elongate, with

exceedingly long verticils. Head with vertex dull black, the posterior orbits broadly obscure yellow; anterior vertex relatively broad, exceeding the diameter of the enlarged antennal pedicel.

Pronotum and cervixal region dark brown. Mesonotal præscutum cinnamon brown, darker brown medially, especially in front; scutal lobes brown; scutellum testaceous, with abundant very long setæ, much longer than those of the præscutal interspaces; postnotum more yellowed. Pleura with propleura and anterior mesepisternum dark liver brown, posterior sclerites more cinnamon brown. Halteres with stem obscure yellow, knob dark brown. Legs with fore coxæ dark brown, remaining coxæ and trochanters brownish yellow; remainder of legs black, tarsal segments two to four snowy white, including the vestiture; femora with erect spinoid setæ on proximal third, tibiæ with similar more scattered setæ over the entire length. Wings with a brown tinge, costal border, including cells C, Sc and outer end of R_1, infuscated, the color continued outwardly as an apical seam over outer end of cell R_3; very narrow and inconspicuous brown seams at origin of Rs, cord and as a cubital seam; prearcular cells concolorous with the ground; veins brown. Veins beyond cord with macrotrichia, including also most of Rs, lacking on both anal veins. Venation: Rs and R_{2+3+4} oblique at origin; cell 1st M_2 about equal to the longest vein beyond it; m-cu at near one-third the length of M_{3+4}; proximal half of cell 2nd A relatively broad.

Abdomen conspicuously bicolored, brownish black, the bases of the segments broadly yellow, including slightly less than one-half of each involving segments two through seven, the eighth telescoped beneath the seventh; hypopygium brownish yellow.

Habitat.—West Papua.

Holotype, male, Araucaria Camp, 800 meters, April 28, 1939 (*Toxopeus*); Alexander Collection.

The only other regional member of the subgenus having white tarsi is *Elephantomyia* (*Elephantomyodes*) *percuneata* Alexander, of the Wisselmeren area of West Papua, a smaller fly with the coloration distinct and with cell 2nd A of the wings very long and narrow.

ERIOPTERINI

TRENTEPOHLIA (MONGOMA) AMISSA sp. nov. Plate 1, fig. 13.

Thorax almost uniformly fulvous yellow; head brownish gray; abdominal tergites orange yellow, the posterior borders of the

intermediate tergites brown; wings pale yellow; cell R_3 very large, cell M_3 open by the atrophy of m.

Female.—Length, about 6.5 millimeters; wing, 7.

Rostrum fulvous; palpi yellow. Antennæ with proximal segments brownish yellow, the outer ones slightly darker brown; flagellar segments long-subcylindrical, slightly longer than the verticils. Head brownish gray, front and the very narrow carinate anterior vertex lighter gray.

Thoracic dorsum almost uniformly fulvous yellow, cervical region, pronotum and anterior præscutum paler yellow. Pleura yellow. Halteres yellow. Legs with coxæ pale yellow; remainder of legs whitened (broken beyond midlength of tibiæ). Wings (Plate 1, fig. 13) pale yellow, unpatterned; veins deeper yellow. Veins glabrous; distal section of R_5 with about seven or eight widely scattered trichia over the whole length. Venation Sc_1 ending about opposite R_2, Sc_2 not evident; R_2 oblique, longer than R_{3+4}; cell R_1 very large; cell M_2 open by atrophy of m; cell R_5 about one-half longer than its petiole, cell M_3 subequal to this element; m-cu shortly beyond fork of M; apical fusion of Cu_1 and 1st A about one-fifth r-m; cell 2nd A broad.

Abdominal tergites orange yellow, posterior borders of the segments narrowly brown, especially distinct on segments four to six; sternites yellow. Ovipositor with cerci strongly upcurved to the acute tips.

Habitat.—West Papua.

Holotype, female, Lower Mist Camp, 1,600 meters, January 15, 1939 (*Toxopeus*); Alexander Collection.

The unique type specimen is badly damaged but the species is so distinct that it is advisable to describe it. In the open cell M_2 the venation introduces still another type to those hitherto known from the Oriental-Australasian fauna. The venation is about intermediate between that in the subgenera *Mongoma* and *Trentepohlia*, and the condition of the medial field in the latter group is produced by a comparable atrophy of the basal section of M_3, as well as m.

TRENTEPOHLIA (MONGOMA) AUSTRALASIÆ Skuse.

Trentepohlia australasiæ Skuse, Proc. Linn. Soc. New South Wales (2) 4 (1890) 834-835, pl. 22, fig. 17 (wing).

Mongoma australasiæ Kertész, Cat. Dipt. 2 (1902) 216.

West Papua: Rattan Camp, 1,200 meters, March 4, 1939 (*Toxopeus*).

TRENTEPOHLIA (MONGOMA) IBELENSIS sp. nov. Plate 1, figs. 14, 16.

General coloration of thorax brownish yellow; rostrum and labial palpi yellow, maxillary palpi and antennæ black; femora and tibiæ dark brown, the extreme tips of the latter whitened, tarsi white; femora with about five basal spinoid setæ, posterior basitarsus with a single strong modified seta; wings tinged with brown, the apex more saturated; R_2 from four to five times R_{3+4}; cell 1st M_2 shorter than vein M_4.

Male.—Length, about 7 millimeters; wing, 7.5.

Rostrum yellow, labial palpi brownish yellow, maxillary palpi black. Antennæ black; flagellar segments long-oval, slightly exceeding the verticils. Head dark liver brown; vertex carinate.

Cervical region and pronotum brownish yellow, the latter with unusually long erect setæ. Mesonotum almost uniformly brownish yellow, with long erect setæ, including series on the præscutal interspaces, sternal setæ elongate. Halteres with stem testaceous yellow, knob infuscated. Legs with coxæ brownish yellow, fore pair slightly darker; trochanters obscure yellow; femora uniformly dark brown, tibiæ similar, the extreme apex whitened; tarsi whitened, proximal end of basitarsus vaguely darkened; posterior femur with about five blackened spinoid setæ, about equally spaced; extreme proximal end of basitarsus dilated, with a single strong seta, followed by a concentration of short subappressed setæ. Remaining legs detached but apparently with the same type of setal armature. Wings (Plate 1, fig. 14) tinged with brown, apex more saturated; stigma darker, small and inconspicuous; veins brown. Macrotrichia on entire length of distal section of R_5. Venation: Sc_1 shorter than vein R_2, the latter about four or five times R_{3+4}; cell 1st M_2 shorter than vein M_4; m-cu at or just beyond fork of M; apical fusion of Cu_1 and 1st A about one-fourth m-cu.

Abdomen brown, the sternites and proximal tergites more yellowed.

Habitat.—West Papua.

Holotype, male, Ibèlè Camp, 2,250 meters, November 27, 1938 (*Toxopeus*); Alexander Collection.

Trentepohlia (*Mongoma*) *ibelensis* is generally similar to *T.* (*M.*) *nigrescens* Alexander, of West Papua and the Territory of New Guinea, especially in the venation. In *nigrescens* (Plate 1, fig. 16) vein R_2 is close to or virtually at the radial fork, greatly shortening vein R_{3+4}, Sc_1 is subequal in length to vein R_2 and cell 1st M_2 is almost equal in length to vein M_4. The

armature of the femora in the two species is different, *nigrescens* being without spinoid setæ.

TRENTEPOHLIA (MONGOMA) MACROTRICHIA sp. nov. Plate 1, fig. 15; Plate 3, fig. 31.

Size medium (wing of female 7.8 millimeters); general coloration of thorax liver brown; femora brown, tibiæ dark brown, tips whitened, tarsi white; fore femora with nearly a score of small spinoid setæ on proximal half, middle and hind femora with about ten longer basal spinoid setæ; proximal end of posterior basitarsus with three black bristles, the outermost very large; wings strongly tinged with brown; vein R_2 about five times R_{3+4}, cell 1st M_2 shorter than vein M_4; abdomen with cerci slender, strongly upcurved to the acute tips.

Female.—Length, about 8.5 millimeters; wing, 7.8.

Rostrum and maxillary palpi dark brown, labial palpi paler. Antennæ with scape and pedicel liver brown, flagellum paler brown; basal flagellar segments long-oval, outer ones more elongate, terminal segment long and slender; verticils shorter than the segments. Head black; vertex carinate.

Cervical region brownish black. Pronotum dark brown, with long erect black setæ. Mesothorax liver brown; præscutum with sparse erect setæ, the posterior ones longer; Halteres dusky. Legs with coxæ liver brown, fore pair darker; trochanters testaceous yellow; femora brown, bases of fore and hind pairs paler; tibiæ dark brown, tips paling to white; tarsi whitened; fore femora with nearly a score of scattered small spinoid setæ on proximal half, middle and hind femora with ten or eleven slightly longer setæ that are restricted to the bases; proximal end of posterior basitarsus with three unequal intensely black setæ, the basal one smallest, outer bristle very long and conspicuous. Wings (Plate 1, fig. 15) strongly tinged with brown, costal border more saturated; narrow vaguely indicated darkened seams over cord, R_2 and vicinity, and at wing tip. Distal section of vein R_5 with about 16 trichia distributed over the entire length. Venation: R_2 from four to five times R_{3+4}; cell 1st M_2 relatively small, shorter than vein R_4; m-cu close to fork of M; apical fusion of Cu_1 and 1st A short.

Abdominal tergites and genital shield dark brown, sternites yellowish brown. Ovipositor (Plate 3, fig. 31) with cercus relatively short, strongly upcurved to the acute tip; genital shield with very long erect to slightly reclinate setæ, some as

long as the cercus. In *tenuicercus* these setæ are pale, shorter than the cerci.

Habitat.—West Papua.

Holotype, female, Lower Mist Camp, 1,550 meters, February 1, 1939 (*Toxopeus*); Alexander Collection.

Trentepohlia (*Mongoma*) *macrotrichia* is best distinguished from other generally similar regional species by the coloration and armature of the legs and the structure of the cerci.

TRENTEPOHLIA (MONGOMA) PARALLELA ALEXANDER. **Plate 1, fig. 17.**

Trentepohlia (*Mongoma*) *parallela* ALEXANDER, Ann. Mag. Nat. Hist. (12) 4 (1951) 598–599.

The unique type was from Kokoda, Territory of Papua, taken by Miss Cheesman. West Papua: Hollandia, July 1, 1938 (*Toxopeus*).

The venation (Plate 1, fig. 17) indicates the distinctness of the species. Particular attention is called to the position of vein R_2 beyond the radial fork, leaving an element R_{2+3} and with veins R_3 and R_4 extending generally parallel to one another, and to the open cubital cell.

TRENTEPOHLIA (MONGOMA) SPINASPERSA sp. nov. **Plate 1, fig. 18.**

Size medium (wing of male 9 millimeters); general coloration dark liver brown; posterior femora with about eight basal spinoid setæ, with fewer widely scattered outer ones, the last at near midlength of segment; wings infuscated, costal border and the very small stigma a little darker; costal fringe short; veins R_2 and R_{3+4} subequal, Sc_1 longer; cell 1st M_2 shorter than vein M_4.

Male.—Length, about 8 millimeters; wing, 9.

Rostrum light brown, labial palpi a little paler, maxillary palpi black. Antennæ with scape and pedicel dark brown, flagellum slightly paler; flageller segments elongate, exceeding the verticils. Head brownish black.

Cervical region and pronotum dark brown. Mesonotum paler liver brown, postnotum more yellowed, especially behind. Pleura dark brown, the posterior parts more yellowed. Halteres infuscated. Legs with fore coxæ dark brown, remaining coxæ and trochanters paler brown; remainder of legs uniformly dark brown, tarsal segments very slightly paler, more so outwardly; posterior femora with eight basal spinoid setæ, the proximal ones longer, with about six additional such setæ

beyond, these more scattered, the last at near midlength of femur; posterior basitarsus with a single poorly differentiated enlarged bristle, not blackened as in some allied species. Wings (Plate 1, fig. 18) infuscated, costal border and the very small stigma a little darker; extreme wing tip slightly infuscated; veins light brown. Costal fringe short; about a dozen trichia scattered over the entire length of distal section of R_5. Venation; Sc_1 very long, exceeding R_2, the latter subequal to R_{3+4}; cell 1st M_2 shorter than vein M_4; m-cu about one-fourth its length beyond fork of M; apical fusion of Cu_1 and 1st A short.

Abdomen dark brown, basal, sternites trifle paler; posterior borders of outer sternites slightly darker.

Habitat.—West Papua.

Holotype, male, Moss Forest Camp, near Lake Habbema, 2,800 meters, October 13, 1938 (*Toxopeus*); Alexander Collection.

In its general appearance, including the uniformly darkened femora and tibiæ, *Trentepohlia* (*Mongoma*) *spinaspersa* suggests *T.* (*M.*) *fimbricosta* Alexander, differing especially in the short costal fringe, details of venation, and specially the armature of the posterior femora.

TRENTEPOHLIA (MONGOMA) SUBAPPRESSA sp. nov. Plate 1, fig. 19; Plate 3, fig. 32.

Size medium (wing 8.8 millimeters); general coloration of thorax liver brown; head black; legs dark brown, tarsi and tips of tibiæ paling to brownish yellow; femora without spinoid setæ, modified bristles at proximal end of posterior basitarsus relatively small and weak; wings strongly tinged with brown; R_2 subequal to or shorter than R_{3+4}; very short to obliterated by approximation of fusion of vein M_{1+2} and M_3; ovipositor with cerci relatively short, compressed-flattened.

Female.—Length, about 8.5 millimeters; wing, 8.8.

Rostrum light brown; labial palpi horn yellow, maxillary palpi black. Antennæ black; flagellar segments elongate, much exceeding the verticles. Head black; setæ conspicuous, mostly slightly proclinate.

Cervical region and pronotum black, setæ of latter very long. Mesonotum liver brown; setæ small and inconspicuous, on anterior end of præscutum. Pleura liver brown. Halteres infuscated. Legs with coxæ liver brown; trochanters brownish yellow; femora dark brown; tibiæ brown, brownish yellow out-

wardly, tarsi brownish yellow; femora without spinoid armature; posterior femur with modified setæ relatively small and weak, scarcely differentiated from the normal bristles. Wings (Plate 1, fig. 19) strongly tinged with brown, costal field and the vaguely indicated stigma slightly darker brown; veins delicate, light brown. About eight scattered macrotrichia on distal section of vein R_5, almost all on outer half. Venation: R_2 subequal to or somewhat shorter than R_{3+4}; vein R_3 oblique; cell 1st M_2 shorter than vein M_4 beyond it, m-cu shortly beyond base; m very short to lacking (as shown) by approximation or short fusion of vens M_{1+2} and M_3.

Abdomen dark brown. Ovipositor with cercus (Plate 3, fig. 32) relatively short and broad, compressed-flattened, upcurved to the narrowly obtuse tip.

Habitat.—West Papua.

Holotype, female, Moss Forest Camp, near Lake Habbema, 2,800 meters, October 31, 1938 (*Toxopeus*); Alexander Collection.

The most similar regional species include *Trentepohlia* (*Mongoma*) *spinaspersa* sp. nov. and *T.* (*M.*) *tenuicercus* sp. nov., best distinguished by the armature of the legs and the structure of the cerci.

TRENTEPOHLIA (MONGOMA) TENUICERCUS sp. nov. Plate 1, fig. 20; Plate 3, fig. 33.

Size medium (wing of female 8.6 millimeters); general coloration of thorax liver brown; femora dark brown, the extreme tips vaguely pale, tibiæ brown, tips more whitened, tarsi dirty white; spinoid armature of femoral bases scarcely developed; R_{3+4} longer than R_2; outer section of M_{1+2} short, about one-third m; apical fusion of Cu_1 and 1st A relative long; ovipositor with cerci long and slender.

Female.—Length, about 10 millimeters; wing, 8.6.

Rostrum dark brown, labial palpi more yellowed, maxillary palpi black. Antennæ black; flagellar segments elongate, subcylindrical, exceeding the verticils. Head brownish black; vertex carinate.

Pronotum liver brown; setæ long and conspicuous. Mesonotum almost uniformly liver brown, setæ shorter and sparse, on præscutum more concentrated at anterior end; setæ of scutellum short, slightly reclinate, sternopleurite with longer setæ. Halteres infuscated, base of stem paler. Legs with coxæ light brown, trochanters paler; remainder of legs elongate, femora

dark brown, extreme tip vaguely pale; tibiæ brown, apex very narrowly more whitened; tarsi dirty white; spinoid setæ of femur lacking or scarcely modified; setæ at proximal end of basitarsus inconspicuous, not enlarged or blackened, the large only about one-half longer than the more distal normal setæ. Wings (Plate 1, fig. 20) tinged with brown, wing tip more suffused; stigma small, darker brown; very narrow brown seams at origin of Rs, base of cell 1st M_2 and on the posterior cord; veins light brown. Macrotrichia over the whole length of distal section of vein R_5. Venation: R_{3+4} longer than R_2; outer elements of cord in oblique alignment; outer section of $_{1+1}$ short, about one-third m; m-cu at or shortly before fork of M, varying slightly in the two wings of the type; apical fusion of Cu_1 and 1st A relatively long, exceeding one-half the length of m-cu.

Abdomen liver brown, the pattern discolored by enclosed eggs. Ovipositor with cercus (Plate 3, fig. 33) unusually long and slender, gently upcurved to the narrowly obtuse tip.

Habitat.—West Papua.

Holotype, female, Top Camp, 2,100 meters, January 25, 1939 (*Toxopeus*); Alexander Collection.

Trentepohlia (*Mongoma*) *tenuicercus* is most similar to species such as *T.* (*M.*) *brevipes* Alexander, *T.* (*M.*) *spinaspersa* sp. nov., *T.* (*M.*) *subappressa* sp. nov. and some others, being best, distinguished by the venation, armature of the legs, and conformation of the cercus of the ovipositor.

GONOMYIA (GONOMYIA) IRIANENSIS sp. nov. Plate 3, figs. 29, 30.

Size medium (wing of male about 6 millimeters); mesonotal præscutum and scutal lobes dark brown, scutellum yellow, pleura yellow, variegated by pale brown; rostrum and halteres yellow; wings milky subhyaline, veins brownish yellow, Sc long; male hypopygium having the inner dististyle with a powerful black curved spine at base; apex of phallosome produced into a long recurved spine.

Male.—Length, about 5.5 millimeters; wing, 6.2.

Rostrum obscure yellow; palpi black. Antennæ black, outer flagellar segments paler; basal flagellar segments long-oval, the outer ones becoming longer and more slender, exceeding their verticils. Head dark gray.

Pronotum and pretergites light yellow. Mesonotal præscutum dark brown, humeral region and lateral borders yellowed;

scutal lobes dark brown, median region more pruinose; scutellum yellow; mediotergite light brown, pruinose, pleurotergite yellowed. Pleura yellow, variegated with pale brown on anepisternum, ventral sternopleurite and meron. Halteres pale yellow. Legs with coxæ and trochanters yellow, fore coxæ somewhat darker; remainder of legs yellowish brown, outer tarsal segments darker. Wings (Plate 3, fig. 29) milky subhyaline, prearcular and costal regions light yellow; veins pale brownish yellow, trichia dark. Macrotrichia on veins of about the outer three-fourths of wing, including about the outer two-thirds of both anal veins. Venation: Sc long, Sc_1 ending about opposite one-third Rs, Sc_2 faint, not far removed; cell R_3 at margin very extensive; basal section of R_5 short; m-cu just beyond fork of M.

Abdominal tergites brown, sternites and hypopygium more yellowed. Male hypopygium (Plate 3, fig. 30) with outer lobe of basistyle, *b*, oval, shorter than the longest setæ. Outer dististyle, *d*, pale, inner apical margin with a glabrous flange; inner style with the two apical fasciculate bristles large, yellow; outer margin at base with a powerful curved blackened spine that bears a single strong seta at near midlength; a small darkened spine on body of style before apex. Phallosome, *p*, pale, with a single blackened apophysis, with a further small lateral spine at near midlength; apex of ædeagus a long recurved spine.

Habitat.—West Papua.

Holotype, male, Moss Forest Camp, near Lake Habbema, 2,850 meters, August 23, 1938 (*Toxopeus*); Alexander Collection.

This is the first record of a member of the typical subgenus from New Guinea. The fly is best distinguished from other species of similar appearance by the hypopygial structure, particularly the inner dististyle and phallosome.

STYRINGOMYIA FUMITERGATA sp. nov. Plate 3, fig. 34.

Allied to *didyma* in structure of the hypopygial dististyle; general coloration of notum brown, patterned with darker, pleura yellowed; hypopygium with tergal lobe narrowed, apex of sternite deeply emarginate; dististyle without a slender outer arm.

Male.—Length, about 7 millimeters; wing, 5.

Rostrum and palpi brown. Antennæ with scape and pedicel blackened, flagellum testaceous yellow. Head brown, setæ un-

usually numerous, including several small yellow bristles additional to the four major ones.

Pronotum brownish yellow, patterned with brownish black, including the lateral borders. Mesonotum brownish yellow, conspicuously patterned with dark brown, including intermediate stripes on anterior half of præscutum, additional to the lateral borders; scutal lobes brown, darker laterally; scutellum brownish yellow, basal depressions conspicuous; postnotum darkened. Pleura chiefly testaceous yellow. Halteres broken. Legs with coxæ and trochanters testaceous yellow; femora yellow, darkened rings narrow but conspicuous, dark brown, narrowly interrupted beneath; tibiæ yellow, tips infuscated, with a further narrow ring just before midlength; tarsi pale, tips of segments very narrowly darkened, terminal segment black. Wings yellowed, with the usual darkened pattern at r-m, outer end of cell 1st M_2, m-cu and less evidently at tip of 2nd A; veins yellow, somewhat darker near margins. Venation: Cell 2nd M_2 narrowly petiolate; vein 2nd A curved strongly to margin.

Abdominal tergites almost uniformly dark brown, with a pale area on either side at and near midlength, sternites and hypopygium more yellowed. Male hypopygium (Plate 3, fig. 34) with the tergite, *t*, narrowed outwardly, apex prolonged into a small relatively narrow setuliferous lobe; disk immediately cephalad of the extended part of lobe with a linear pocket of dense yellow setæ. Sternite, *s*, long and narrow, apex with a large U-shaped emargination, lateral lobes very slender, each tipped with a long black bristle that is about one-third longer than the lobe; surface with abundant delicate setulæ and a few normal setæ. Basistyle, *b*, with a single spinoid seta, unusually stout, subequal in length to its basal lobe. Dististyle, *d*, unusually compact, without an elongate outer arm, the conformation about as figured, including two flattened blades that are margined with black spinoid setæ, the larger blade with abundant long setæ on outer margin; a small slender blade, tipped with an acute spine. Phallosome, *p*, simple, unusually pale, the only blackened part being a small apical point.

Habitat.—West Papua.

Holotype, male, Sigi Camp, along Sigi River, 1,500 meters, February 26, 1939 (*Toxopeus*); Alexander Collection.

Styringomyia fumitergata is allied to *S. biroi* Edwards, *S. didyma* (Grünberg), and *S. kertészi* Edwards, all differing

among themselves in coloration, particularly of the abdomen, and in the details of structure of the male hypopygium.

STYRINGOMYIA MULTISETOSA sp. nov. Plate 3, fig. 35.

Allied to *ensifera;* male hypopygium with apical tergal lobe very long, with abundant retrorse yellow setæ; sternite broad, with several strong subapical setæ; basistyle with two modified spinoid setæ; dististyle complex, all three arms with combs of blackened spinoid setæ.

Male.—Length, about 7 millimeters; wing, 6.

Female.—Length, about 6 millimeters; wing, 5.

Rostrum and palpi brownish yellow. Antennæ with scape darkened, remainder of organ clearer yellow. Head light brown; vertex with four powerful black bristles, two on the anterior vertex more divergent, the posterior pair more correct.

Thorax obscure yellow, faintly patterned with darker; præscutal setæ relatively weak; each scutal lobe with a single strong erect to slightly porrect bristle; mediotergite with a pair of more slender erect setæ. Pleura yellow, clearer ventrally. Halteres obscure yellow. Legs with coxæ and trochanters light yellow; femora yellow, each with two brown rings that are much narrower than the interspace; tibiæ and tarsi yellow, terminal tarsal segment black. Wings light yellow, with small brown spots at r-m, m-cu, and outer end of cell 1st M_2; small marginal darkenings from R_5 to 2nd A, the last large. Venation: Cell 2nd M_2 sessile; vein 2nd A unspurred.

Abdomen yellow, tergites conspicuously patterned with brown, including a pair of spots in basal half and the posterior border, these marks more extensive and crowded on outer segments. Male hypopygium (Plate 3, fig. 35) with the apical lobe of tergite, *t*, very long, narrowed to the obtuse tip, surface with very abundant retrorse yellow setæ. Sternite, *s*, broad, apex with several strong setæ, the larger ones subterminal. Basistyle, *b*, with two modified spinoid setæ, the outer one more slender, sessile; inner seta from a low tubercle, its tip less produced. Dististyle, *d*, complex in structure, as shown; outer arm with the slender outer half darkened, base dilated, its outer end with numerous delicate setæ, base of arm with two combs of spinoid setæ, the linear basal ones shorter; intermediate and inner arms of style as shown, each with a comb of spinoid setæ; inner arm with additional armature, as shown, the outer spine slender; at base of style with a further slender arm with a recurved blackened spine, its tip obtuse. Phallosme, *p*, massive, as shown.

Habitat.—West Papua.

Holotype, male, Rattan Camp, 1,200 meters, February 12, 1939 (*Toxopeus*); Alexander Collection.

Other related regional species having the basistyle of the hypopygium bispinous include *Styringomyia ensifera* Edwards and *S. ensiferoides* Alexander, all three species differing among themselves in hypopygial characters, particularly the tergite, sternite and dististyle, especially the intermediate and inner arms of the last.

STYRINGOMYIA PHALLOSOMICA sp. nov. Plate 3, fig. 36.

Allied to *spinicaudata;* general coloration yellow, mesonotum patterned with black; legs yellow, darkened rings of femora and tibiæ pale and inconspicuous; wings yellow, faintly patterned with brown, appearing chiefly as narrow seams and darkenings to the veins; male hypopygium with tergite broad, outer apical angles slightly produced; apex of sternite produced medially; all arms of dististyle with conspicuous spinoid setæ; phallosome with a single terminal spine and two pairs of marginal projections, the outer ones acute.

Male.—Length, about 6.8 to 7 millimeters; wing, 5 to 5.2.

Rostrum and palpi brownish yellow. Antennæ with scape light brownish yellow, slightly darkened beneath; pedicel brownish yellow; flagellum light yellow, outer segments slightly darkened. Head yellow, anterior vertex slightly darker; two pairs of setæ on anterior vertex, the longer anterior pair divergent, setæ of posterior vertex weak.

Pronotum above light brown, sides yellow. Mesonotum yellow, patterned with black including a Ω-shaped area on either side of præscutum, with further darkenings on scutal lobes; sides of scutellum infuscated, postnotum darkened. Pleura and pleurotergite light yellow. Halteres yellow. Legs with coxæ and trochanters yellow; remainder of legs yellow, femoral rings very pale brown, inconspicuous, tibial darkenings more evident; terminal tarsal segment black; setæ of posterior legs longer, especially on femora; outer ends of femora, tibiæ and basitarsi with scattered blackened bristles. Wings yellow, faintly patterned with brown, including narrow seams in cord, outer end of cell 1st M_2, m-cu, distal end of vein Cu and less evidently at tip of 2nd A, the markings restricted and indicated chiefly by darkening of the veins which otherwise are light yellow. Sparse macrotrichia on both branches of R, lacking on M. Venation: Cell 2nd M_2 at base with petiole punctiform or lacking; vein 2nd A simple.

Abdomen conspicuously bicolored, yellow, posterior borders broadly dark brown. Male hypopygium (Plate 3, fig. 36) with the tergite, *t*, broad, posterior border subtruncate to very gently convex, outer lateral angles slightly produced; apical margin, including the lobes, with longer setæ, those of the disk exceedingly minute. Sternite, *s*, narrowed outwardly, the central part farther produced; glabrous or with a few microscopic setulæ only; outer end of sternite with long setæ. Basistyle, *b*, with a single long spinoid seta, this about two and one-half to three times as long as its basal tubercle. Dististyle, *d*, with outer arm delated, with long retrorse setæ throughout its length, terminal elongate bristle normal; intermediate arm flattened, provided with long black setæ, about as figured; inner arm more slender, the spinoid setæ shorter. Phallosome, *p*, distinctive, as figured, including an unpaired outer spine and two pairs of blackened points, the outer pair acutely pointed, the basal ones obtuse.

Habitat.—West Papua.

Holotype, male, Lower Mist Camp, 1,600 meters, January 15, 1939 (*Toxopeus*). Paratopotype, male.

Styringomyia phallosomica is allied to *S. platystyla* Alexander and *S. spinicaudata* Alexander, especially to the former, in the patterned wings and general structure of the hypopygium, differing particularly in the various details of the latter, including the tergite, sternite, dististyle and phallosome.

STYRINGOMYIA SETIFERA sp. nov. Plate 3, fig. 37.

Allied to *biroi;* size very small (wing less than 4 millimeters); general coloration yellow, patterned with darker; male hypopygium with apex of tergite rounded, without lateral shoulders; apex of sternite emarginate; dististyle with only two sets of lobes, variously modified, as described.

Male.—Length, about 5.5 millimeters; wing, 3.8.

Female.—Length, about 4 millimeters; wing, 3.4.

Rostrum light yellow, palpi darker. Antennæ with scape blackened beneath, light yellow above, pedicel black, flagellum obscure yellow. Head buffy yellow.

Pronotum with disk yellow, margins dark brown. Mesonotal præscutum light yellow, the cephalic lobe dark brown, disk with the interspaces weakly infuscated, setæ relatively short; laterally behind with a darkened area that crosses the suture and covers the lateral parts of the scutal lobes; a single erect seta on mesal

edge of each scutal lobe; scutellum pale yellow, with two erect setæ, parascutella slightly darker; mediotergite brownish yellow. Pleurotergite and pleura reddish yellow. Halteres pale yellow. Legs with coxæ reddish yellow; trochanters yellow; femora yellow, with two narrow and incomplete darkened spots; tibiæ with apex and a weak ring before midlength darkened, the setæ at these points stouter and blackened; tarsi obscure yellow, terminal segment black; posterior femora of male on outer two-thirds with a series of more than a dozen very long erect setæ that are subequally spaced. Wings pale yellow, with the usual four darkened spots, placed at r-m, outer end of cell 1st M_2, m-cu and tip of vein 2nd A. Venation: Cell 2nd M_2 narrowly sessile; vein 2nd A strongly curved to margin, unspurred.

Abdomen yellow, posterior tergal borders narrowly darkened, the areas notched but not completely interrupted medially; seventh tergite with a median dark T-shaped area. Male hypopygium (Plate 8, fig. 37) with apical lobe of tergite, *t*, rounded, without lateral shoulders; setæ directed backwards, on disk with a triangular group of longer yellow setæ directed cephalad. Sternite, *s*, narrow and pale, apex with a conspicuous U-shaped, notch, lateral lobes each with a single seta about three times its own length. Basistyle, *b*, with a single spinoid seta, a little longer than its basal tubercle, on mesal face of latter at base with a fingerlike spur. Dististyle, *d*, compact, about as shown, without a slender outer arm, as is common in the genus; the usual intermediate lobe flattened and more of less scooplike, the broader lobe with a small lateral spicule; inner arm elongate, the longer part narrowed outwardly, provided with a double row of spinoid setæ; inner part shorter, terminating in a compact group of blackened peglike setæ. Phallosome, *p*, relatively slender, including two outer flattened blades, before their bases with two devergent blackened spines.

Habitat.—West Papua.

Holotype, male, Bernhard Camp, on Idenburg River, 50 meters, September 27, 1938 (*Toxopeus*). Allotopotype, female.

Styringomyia setifera is allied to *S. biroi* Edwards and *S. kertészi* Edwards in the lack of the outer arm of the dististyle of the hypopygium. All three species differ evidently in the details of the hypopygium, particularly the tergite, dististyle and phallosome. Edward's figures of his species are somewhat diagrammatic but clearly indicate that there are two distinct species involved.

TOXORHINA (TOXORHINA) DIGITIFERA sp. nov. Plate 3, fig. 38.

Size small (wing of male 4.3 millimeters); general coloration of thorax black, more or less pruinose; rostrum short, about two-thirds the wing; halteres yellow; wings tinged with brown; macrotrichia on all veins beyond cord, Rs and distal third of M; m-cu beyond fork of M; dististyle of male hypopygium with a slender fingerlike lobe on outer margin between the basal horn and the beak.

Male.—Length, excluding rostrum, about 3.6 millimeters; wing, 4.8; rostrum, about 2.8.

Rostrum black, about two-thirds the wing. Antennæ black throughout. Head large, blackened pruinose.

Mesonotum discolored; præscutum and scutal lobes black, humeral region paler, sides of præscutum light gray pruinose; posterior sclerites of notum brownish black. Pleura brownish black, more pruinose on dorsal sternopleurite and ventral pteropleurite. Halteres yellow. Legs with coxæ brownish yellow, tips clearer yellow; trochanters brownish yellow; femora light yellowish brown but appearing darker because of the vestiture. Wings tinged with brown, in one wing of the type with the outer margin of radial field more suffused, this condition not evident on the opposite wing; veins brown, trichia black. Veins beyond level of origin of Rs with macrotrichia, including all veins beyond cord, Rs, and distal third of M. Venation: Sc short, Sc_1 ending a very short distance beyond origin of Rs, Sc_2 far retracted; cell R_5 narrowed at margin; m-cu longer than distal section of Cu_1 or nearly three times the basal section of M_{3+4}.

Abdomen brownish black, hypopygium slightly more brightened. Male hypopygium (Plate 3, fig. 38) with posterior border of tergite, *t*, convexly rounded, broadly obtuse. Basistyle, *b*, with setæ simple, relatively sparse, those of mesal face small and weak. Dististyle, *d*, with beak very long and narrow, gradually narrowed to the obtuse tip; outer margin with a short curved hornlike lobe, in its axil with a long slender fingerlike lobe, about two-thirds the length of the dilated base, lying generally parallel to the beak; outer dististyle small and weak, sinuous, closely appressed to the base of the major style. Ædeagus, *a*, with arms unusually long and slender, closely appressed to one another.

Habitat.—West Papua.

Holotype, male, Mist Camp, Idenburg River, 1,800 meters, January 4, 1939 (*Toxopeus*); Alexander Collection.

Toxorhina (*Toxorhina*) *digitifera* is quite distinct from other regional members of the subgenus in the structure of the hypopygium, particularly the dististyles.

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ILLUSTRATIONS

[Legend: *a*, aedeagus; *b*, basistyle; *d*, dististyle; *g*, gonapophysis; *p*, phallosome; *s*, sternite; *t*, tergite.]

Plate 1

Fig. 1. *Limonia* (*Laosa*) *pavo* sp.nov.; venation.
2. *Limonia* (*Laosa*) *suffalcata* sp.nov.; venation.
3. *Limonia* (*Libnotes*) *invicta* sp.nov.; venation.
4. *Limonia* (*Libnotes*) *tibiocincta* sp. nov.; venation.
5. *Limonia* (*Rhipidia*) *dione* sp.nov.; venation.
6. *Limonia* (*Dicranomyia*) *eucharis* sp.nov.; venation.
7. *Limonia* (*Dicranomyia*) *niveifusca* sp.nov.; venation.
8. *Limonia* (*Geranomyia*) *atychia* sp.nov.; venation.
9. *Limonia* (*Geranomyia*) *dicranostyla* sp. nov.; venation.
10. *Limonia* (*Thrypticomyia*) *arcus* sp.nov.; venation.
11. *Helius* (*Helius*) *subanacmicus* sp.nov.; venation.
12. *Helius* (*Rhyncholimonia*) *dicroneurus* sp.nov.; venation.
13. *Trentepohlia* (*Mongoma*) *amissa* sp.nov.; venation.
14. *Trentepohlia* (*Mongoma*) *ibelensis* sp.nov.; venation.
15. *Trentepohlia* (*Mongoma*) *macrotrichia* sp.nov.; venation.
16. *Trentepohlia* (*Mongoma*) *nigrescens* Alexander; venation.
17. *Trentepohlia* (*Mongoma*) *parallela* Alexander; venation.
18. *Trentepohlia* (*Mongoma*) *spinaspersa* sp.nov.; venation.
19. *Trentepohlia* (*Mongoma*) *subappressa* sp.nov.; venation.
20. *Trentepohlia* (*Mongoma*) *tenuicercus* sp.nov.; venation.

Plate 2

Fig. 21. *Limonia* (*Libnotes*) *invicta* sp.nov.; male hypopygium.
22. *Limonia* (*Rhipidia*) *dione* sp.nov.; male hypopygium.
23. *Limonia* (*Dicranomyia*) *eucharis* sp.nov.; male hypopygium.
24. *Limonia* (*Dicranomyia*) *niveifusca* sp.nov.; male hypopygium.
25. *Limonia* (*Geranomyia*) *atychia* sp.nov.; male hypopygium.
26. *Limonia* (*Geranomyia*) *dicranostyla* sp. nov.; male hypopygium
27. *Helius* (*Helius*) *subanacmicus* sp.nov.; male hypopygium.
28. *Helius* (*Rhyncholimonia*) *dicroneurus* sp.nov.; head, antenna.

Plate 3

Fig. 29. *Gonomyia* (*Gonomyia*) *irianensis* sp.nov.; venation.
30. *Gonomyia* (*Gonomyia*) *irianensis* sp.nov.; male hypopygium.
31. *Trentepohlia* (*Mongoma*) *macrotrichia* sp.nov.; ovipositor, cercus.
32. *Trentepohlia* (*Mongoma*) *subappressa* sp.nov.; ovipositor, cercus.
33. *Trentepohlia* (*Mongoma*) *tenuicercus* sp.nov.; ovipositor, cercus.
34. *Styringomyia fumitergata* sp.nov.; male hypopygium.
35. *Styringomyia multisetosa* sp.nov.; male hypopyguim.
36. *Styringomyia phallosomica* sp.nov.; male hypopygium.
37. *Styringomyia setifera* sp.nov.; male hypopygium.
38. *Toxorhina* (*Toxorhina*) *digitifera* sp.nov.; male hypopygium.

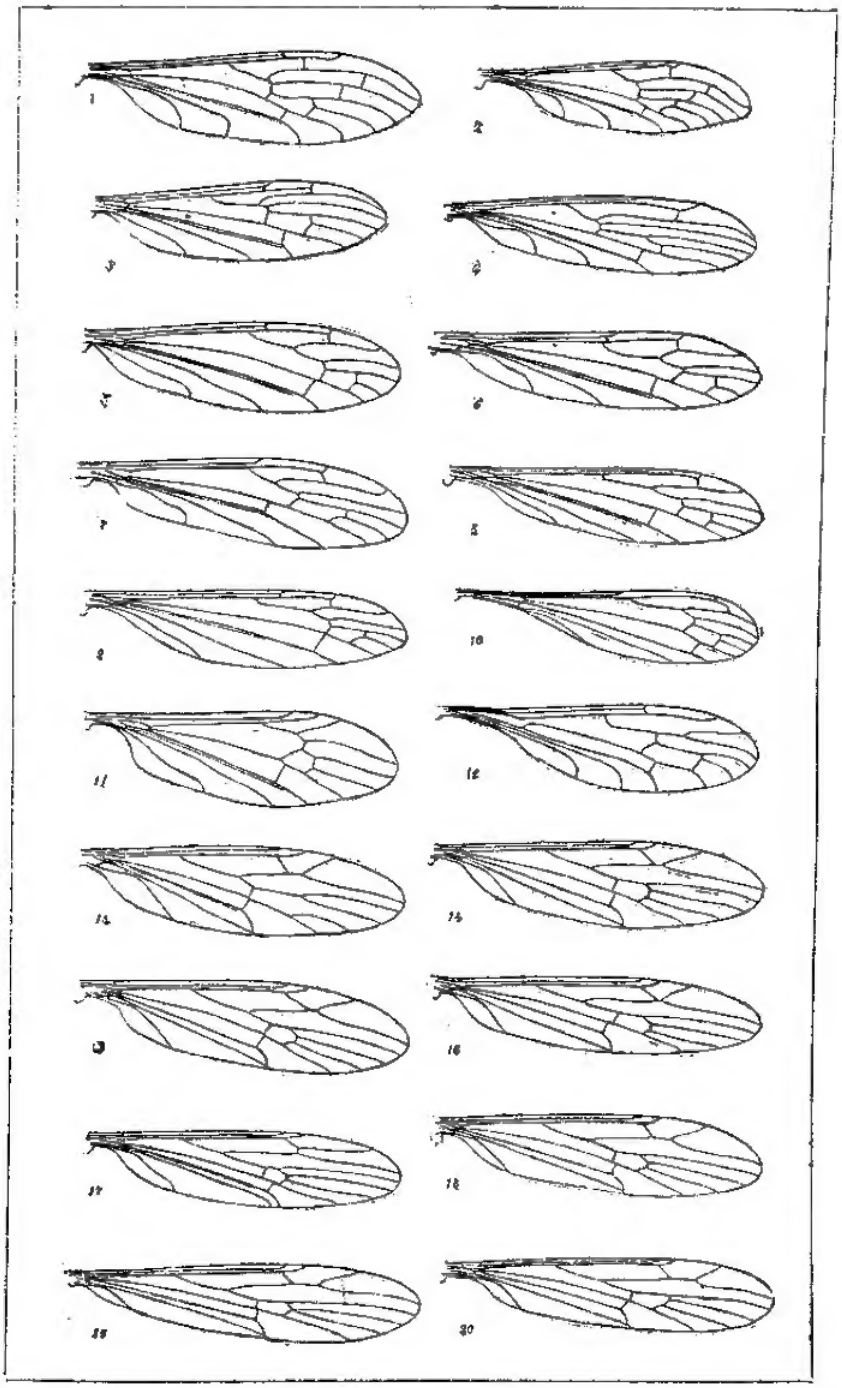

PLATE 1.

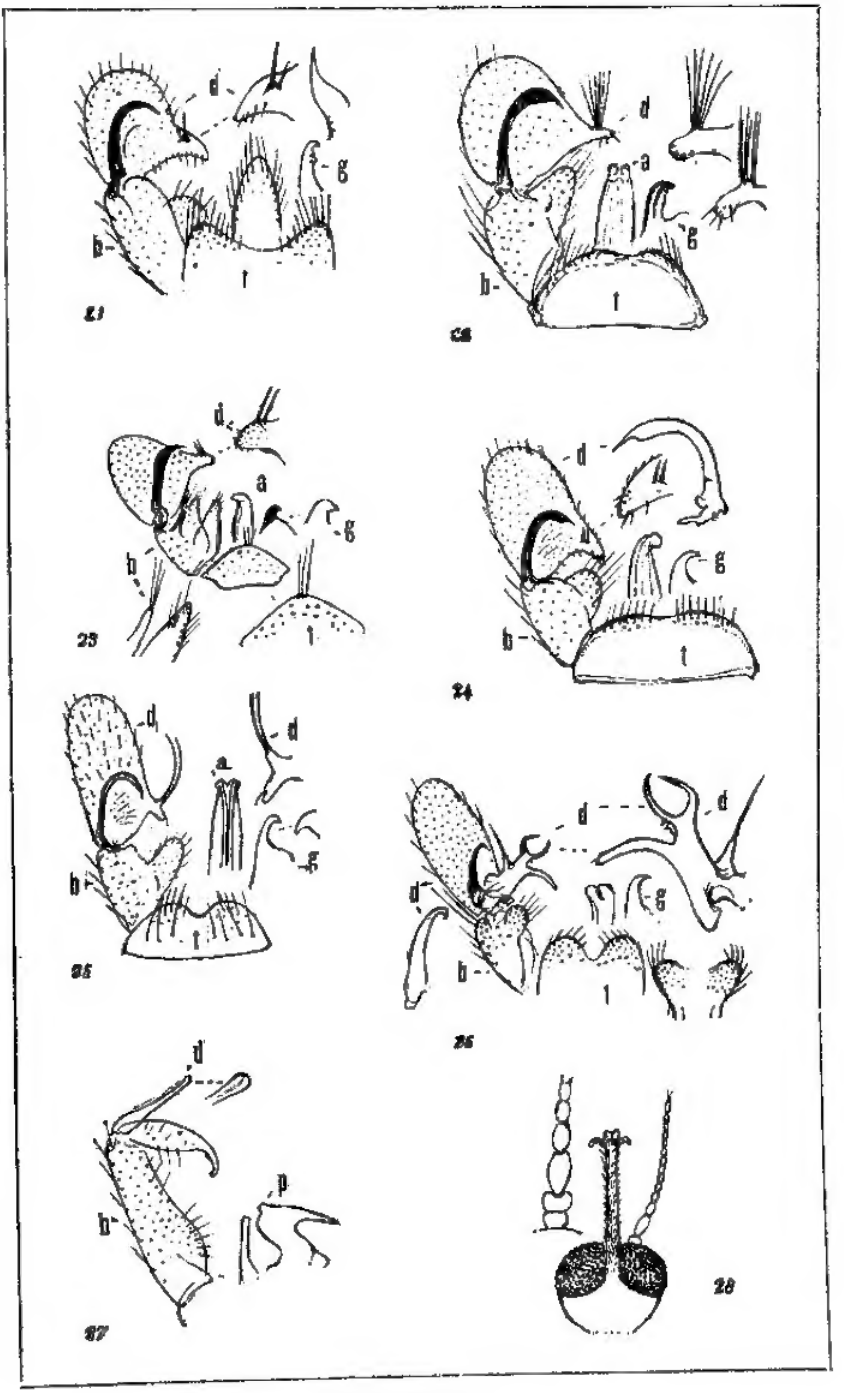

PLATE 2.

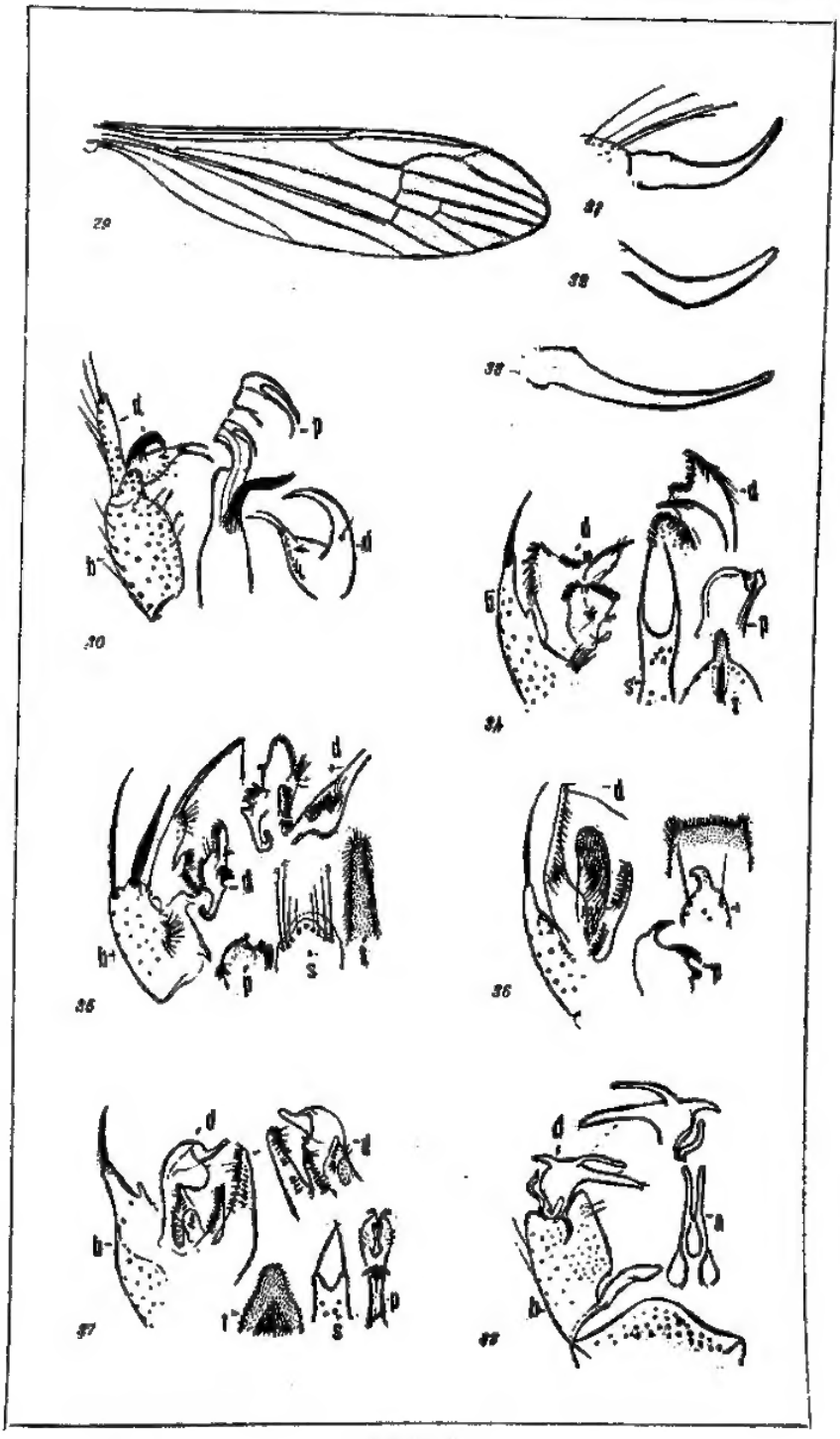

PLATE 3.